Development of a Multiplex PCR Diagnostic Test for Diarrheal Parasites Commonly Found in Jordan

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Background

• The World Health Organization reports that diarrhea causes 1.5 million child deaths per year.
• 13% of Jordanian children under age 5 who died in 2004 were killed by diarrhea (WHO).
• Microscopy is the common method for parasite diagnostics in developing countries, but this technique is not sensitive enough to detect all parasites in a stool sample nor sufficient in distinguishing between different parasites.

Parasitic Diseases

• Cryptosporidium and Giardia parasites cause the severe diarrheal diseases Cryptosporidiosis and Giardiasis, respectively.

Methods

• Primers were designed to anneal at ribosomal subunits. These DNA regions are variable from species to species and sufficiently conserved within a species to allow for amplification specificity.

Results

• MP Primer Computer Program was used for multiplex primer design.

• Each parasite’s specific primers were tested individually on Giardia and Cryptosporidium DNA.

• Gel electrophoresis was used to analyze the PCR product.

Purpose

• We aim to develop a single multiplex PCR test that will distinguish between the genera of Cryptosporidium and Giardia and between the species of Entamoeba histolytica and E. dispar.

Future Experimentation

• PCR with varied combinations of DNA template, primers and adjusted PCR parameters.

• If awarded the Fulbright Fellowship, Hiba Jamil will implement the multiplex PCR test at the Princess Haya Biotechnology Center at the Jordan University of Science and Technology in Irbid, Jordan in September 2011.

• She will be running the assay on samples from immunocompromised patients at the King Hussein Cancer Center in Amman, Jordan.

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