Cryptosporidium IMS Beads: For the isolation and concentration of *Cryptosporidium spp.* oocysts by ImmunoMagnetic Separation (IMS)

PRODUCT INSERT

FOR RESEARCH USE ONLY

Catalog No.: IMS206:Crypto (2 ml)

Introduction:

The Virusys *Cryptosporidium* IMS Beads (Crypto IMS Beads) have been developed for the immunomagnetic separation (IMS) of *Cryptosporidium spp.* oocysts from a wide range of sample matrices. The high sensitivity and specificity of the Crypto IMS Beads results from the covalent attachment of a high affinity monoclonal antibody developed against an epitope on the wall of *Cryptosporidium* oocysts. When combined with the Virusys IMS Assay Buffer (IMS206:BFR) and Virusys IMS Wash Buffer (IMS206:Wash), *Cryptosporidium* oocysts can be efficiently isolated and enumerated by a variety of techniques including, but not limited to immunofluorescent antibody (IFA) staining and PCR.

Intended Use:

The Virusys Crypto IMS Beads are intended for the immunomagnetic separation of *Cryptosporidium spp.* oocysts from a wide range of sample matrices including water sample concentrates.

Virusys Corporation

P.O. Box 56 • Taneytown, MD 21787 Web: www.virusys.com • E-mail: support@virusys.com Virusys Corporation

Procedure:

The Virusys Crypto IMS Beads are supplied ready to use. For most applications, 100 ul of beads will be sufficient for the capture of *Cryptosporidium* oocysts in a final volume of 10 ml. The following procedure should be considered as generic. Each user should anticipate optimizing the system for use in their laboratory, including the utilization of appropriate controls.

1. Transfer the sample to an IMS capture tube in a final volume of 10 ml.

NOTE: IMS Capture Tubes are 16 x 25 mm screwcapped glass tubes with a flat window measuring 11 x 55 mm (Virusys IMS-206:LT).

Virusys recommends the use of one part (5 ml) Virusys IMS Assay Buffer (IMS-206:BFR) and one part (5 ml) reagent water or other diluent for this step (The Virusys IMS Assay Buffer was developed to maximize the capture of a variety of organisms using antibody-coated paramagnetic beads, while minimizing the nonspecific trapping and attachment of materials derived from complex sample matrices).

- Vortex the Crypto IMS Beads for 25-30 sec. and add 100 ul of beads to each IMS capture tube.
- 3. Place the IMS capture tube(s) on a rotating mixer (15-20 rpm) and rotate for 1 hr.
- 4. Remove the tube from the rotating mixer and place on a paramagnetic particle processor with the flat side of the tube toward the magnet.
- 5. Hold the tube/magnet in the horizontal so that the flat side of the tube is <u>above</u> the rounded side of the tube and rock the tube for 3-4 min.
- 6. While maintaining the position of the tube, pour off the supernatant.
- 7. Add 0.5 ml of Virusys IMS Wash Buffer (or other suitable reagent) to the IMS capture tube, remove the tube from the magnet, and gently rinse the beads from the flat surface.
- 8. Transfer the bead suspension to a 1.5-2 ml microcentrifuge tube.
- Add an additional 1 ml of IMS Wash Buffer to the IMS capture tube and place on the magnet. Rinse the IMS capture tube by rocking for 30-60 sec. and collect the residual beads on the magnet.
- 10. Remove the tube from the magnet, resuspend the beads in the Wash Buffer, and transfer the beads to the microcentrifuge tube.

- 11. Place the IMS capture tube in a vertical position and allow to stand for 1 min. Collect and transfer any residual fluid/beads to the microcentrifuge tube.
- 12. Cap and rock the microcentrifuge tube 10-12 times to resuspend all the beads.
- 13. Attach tube to the paramagnetic bead processor. Rock the tube for 1-2 min. Aspirate and discard supernatant.
- 14. If the sample still contains sediment, it may be necessary to rinse the beads with an additional 1 ml of IMS Wash Buffer.

The sample is now ready for additional processing. Depending upon the specific application(s) of the end user, dissociation of the organisms from the beads may be required. Check the Virusys website for more information.

Virusys Corporation