Reagent Preparation Sheet.

Preparation of Phage Lysin digested sample for SeM ELISA

Date prepared:

By:

I. **Hazard Assessment**

To protect yourself from any possible hazards associated with this task wear eye protection. You should also wear latex, nitrile, or vinyl gloves and a lab coat with long sleeves. To protect your legs and feet wear closed shoes and long trowsers. Do not wear sandles, shorts or a short skirt. Wash your hands before eating and when leaving the laboratory. You should review the MSDS for any chemical used in this procedure. In case of a spill with a toxic chemical remove all contaminated clothing and wash affected areas with copious quantities of water. Check location of the nearest safety shower. Eyes should be washed copiously for 15 minutes.

II. **Reagent**
Phage lysin digest of sample for SeM ELISA.

III. **Purpose of reagent**
The Phage Lysin C (PlyC) releases SeM from the wall of *Streptococcus equi* by digesting the peptidoglycan. This exposes epitopes on the SeM protein which are recognized by MAB212p (capture) and MAB211p (detector) monoclonal antibodies.

IV. **Reconstituted vial and storage**
Phage lysin was received at 5 mg/ml (i.e. 5 ug/ul) and aliquotted into 50 ul aliquots and frozen at -20 C.

V. **To digest sample**

The following method is used to digest the sample. The sample is intended to be on a cotton swab. This would be either a clinical swab, a swab placed in compost, a swab swept through the water (or over the plastic surface) of a water bucket, or a swab drawn over a blood plate to harvest streptococcal colonies

Twirl swab in 1 mL of PBS (0.15 M, pH 7.6)	_____
Add 5 ul of phage lysin	_____
Incubate 30 minutes at 37 C	_____
Label, date and store unused digest at - 20 C	_____

VI. **Comments**