Macdonald Campus Farm Cattle Complex
Standard Operating Procedure # DC-616

MILK CULTURING

1. PURPOSE

To detect and treat mammary infections quickly and appropriately in cows with;

- High somatic cell count
- Clinical and sub clinical mammary infections.
- Fresh cows (as early as 7th milking postpartum)

2. RESPONSIBILITY

2.1 Trained and qualified personnel.

3. GENERAL

3.1 Collect aseptic milk samples as per SOP DC-615: Milk Sampling.

3.2 CMT= California mastitis test

4.

4.1 Disinfectant spray for surfaces
4.2 Paper towel
4.3 Dish soap
4.4 Clean dish/container
4.5 Sterile sample tube
4.6 Test tube holder
4.7 Sharpie
4.8 Butterfields buffer dilution tubes (9ml)
4.9 Sterile pipettes or pipette tips
4.10 Micro pipette or Pipette bulb
4.11 Incubator (White foam egg incubator set at 35 degrees C and or CheckUp incubator set at 37 degrees C)
4.12 Spreader
4.13 3M Petrifilm plates: Aerobic (AC), Coliform (CC), Staph Express Count Plate (STX), Staph Express Disk (stored in pharmacy freezer)
4.14 Petridish
4.15 Hand sanitizer
4.16 Checkup Petri dish (kit)

5.1 PREPARATION:

5.1.1 Plug in the incubator(s) you will be using to warm them up.

5.1.2 Disinfect the work surface with spray.

5.1.3 In a clean dish:

5.1.3.1 Add a drop of dish soap to cold/lukewarm water.

5.1.3.2 Insert the milk sample tubes to wash the exterior. This is to minimize contamination of the cultures.

5.1.3.3 Rinse the tubes.

5.1.3.4 Place the tubes in the test tube holder. Separate any quarters that have clinical mastitis or have tested positive on the CMT.

5.1.4 Prepare all other materials needed (buffer, empty sample tube,

5.6.1.2.1 The CC plate spreads itself evenly on its own.

5.6.1.8 AC Petrifilm:

5.6.1.2.1 Using the same pipette tip, inoculate the AC plate and drop the top film down over the sample.

5.6.1.2.1 Turn the spread over to the side with the ridge.

5.6.1.2.1 Place over the center of the inoculated milk and press down to form a perfect circle.

5.6.5 Place all Petrifilm in the incubator. They can be stacked one on top of the other (max 10)

5.6.6 Rinse/wash all equipment and return.

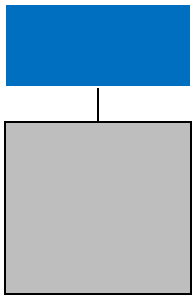
5.6.7 Dispose of used pipette tips and diluted milk. Return bag of 3M Petrifilm to the freezer. Milk samples are frozen in case we need to retest at a later date.

5.6.8 Check the incubator temperature throughout the day. Adjust the temperature by opening or closing the air vent at the top if necessary to maintain at 35°C.

5.6.9 Check the Petrifilm in 24 hours.

5.7 INTERPRETATION OF RESULTS:

5.7.1 STX Petrifilm.



5.7.2 CC Petrifilm:

5.7.3 AC Petrifilm

If interpretation is difficult, or identification of bacterial strain is required, perform a Checkup Petridish Culture. See Section 5.7

5.7.4 Record the observations in the Treatment logbook (cow #, quarter # or pooled sample, dilution, result, # of colonies present)

5.7 CHECKUP PETRI DISH CULTURES: For use when difficult to interpret the results of the 3M Petrifilm and for clinical mastitis.

5.7.1 Plug in the Check Up incubator.

5.7.2 Use aseptic techniques.

5.7.3 Follow the plating protocol as per described on pages 11 and 12 of the Checkup Instruction manual.

5.7.4 Place
Results in 24 hours or more.

5.7.5 Reference the Culture Interpretation Guide section of the instruction manual to interpret the results.

5.7.6 Dispose of bag with Petri dish in the bio box.

5.7.7 Record the observations in the Treatment logbook (cow #, quarter # or pooled sample, dilution, result, # of colonies present).

5.8 Discuss treatment options and with Technician or Herd Manager. Treatment depends