On-Farm Culture Setup

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On-farm culturing can be a valuable tool in mastitis management. Causative bacteria can be identified within 24 to 28 hours for roughly $4 to $5 a sample. Supplies needed before starting on-farm culturing are: 1) a clean and temperature controlled room to use as a laboratory, 2) incubator, 3) bi or tri-culture plates, 4) gloves, 5) a tube rack, 6) sterile cotton swabs, 7) a permanent marker for labeling plates, and 8) a culture guide. A work table or counter top works nicely to arrange all of the necessary equipment to develop a step-wise approach. A simple but inclusive record keeping system is imperative to the success of a culture program. Spreadsheets can be created with Microsoft Excel, which comes with most Microsoft Office software. Results can also be entered into herd management software, such as PCDART.

1. The incubator and work space should be setup in an area that is easy to disinfect and should always be kept clean to prevent any contamination that might contribute to faulty results. The area should be temperature and humidity controlled. The incubator should be set at a temperature of 98.6 F (37 C) and have a container of water in the bottom of the incubator. This allows bacteria to grow in a warm and moist environment.
2. Media plates (bi or tri-culture plates) should be stored in a refrigerator upside down (lid side down with the agar facing down). Do not use expired media plates.
3. Maintain records including the sample date, cow ID, affected quarter (use RF for right front, RR for right rear, LR for left rear, LF for left front) and culture results. Others include type of sample: post-calving, high somatic cell count, clinical signs, etc. Figure 1 shows a sample record sheet.

Sample Date:	Cow ID:	Quarter:	Results:

Figure 1. *Example record sheet (photo credit Kody Hash).*

4. Wear clean gloves at all times. Changing gloves in between samples from different animals can reduce the likelihood of faulty results.
5. Make sure all samples are collected aseptically. Improperly collected samples may cause faulty results. For information about how to properly collect aseptic samples, see Aseptic Milk Sampling Guide (W 945).
6. Fresh samples can be plated immediately. Frozen samples need to be completely thawed before plating. Mix milk tubes well by inverting the sample 10 times. Do not allow samples to sit at room temperature for more than 30 minutes.
7. Start with a new plate and sterile cotton swab. Avoid touching the cotton swab or plate with your fingers or other surfaces as this will result in contamination. Place the plate upside down and label it with the date the sample was taken, the cow it was collected from, and the quarter (RF, RR, LR, or LF) (Figure 2). *Note: It is strongly discouraged to use composite samples from two or more quarters. Using a sample from a single quarter will ensure that you know exactly which quarter to treat.

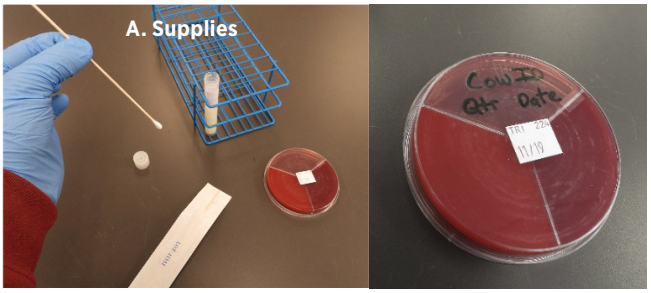

Figure 2. *Example of supplies needed (a) and properly labeling a petri-dish (b) for on-farm culturing (photo credit Kody Hash).*

8. Open the milk sample and put the cotton swab inside. Allow milk to saturate the cotton end for approximately 10 seconds (Figure 3).

9. Pick up the media plate and cover the entire media section with the swab using a side-to-side motion without lifting the swab up (Figure 3 and 4). *Note: Apply gentle pressure to the swab, as you do not want to dig the swab into the media.

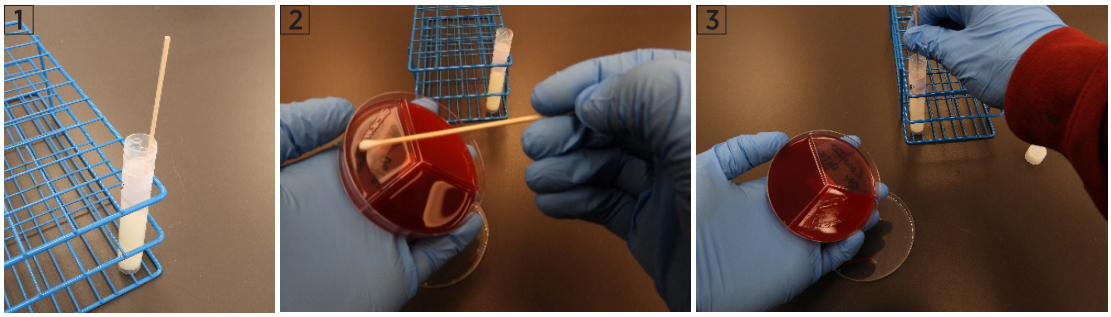

Figure 3. *Step 1) saturate the cotton swab in the milk sample, Step 2) use the cotton swab to apply milk to the media plate, and Step 3) replace the cotton swab in the milk sample (photo credit Kody Hash).*

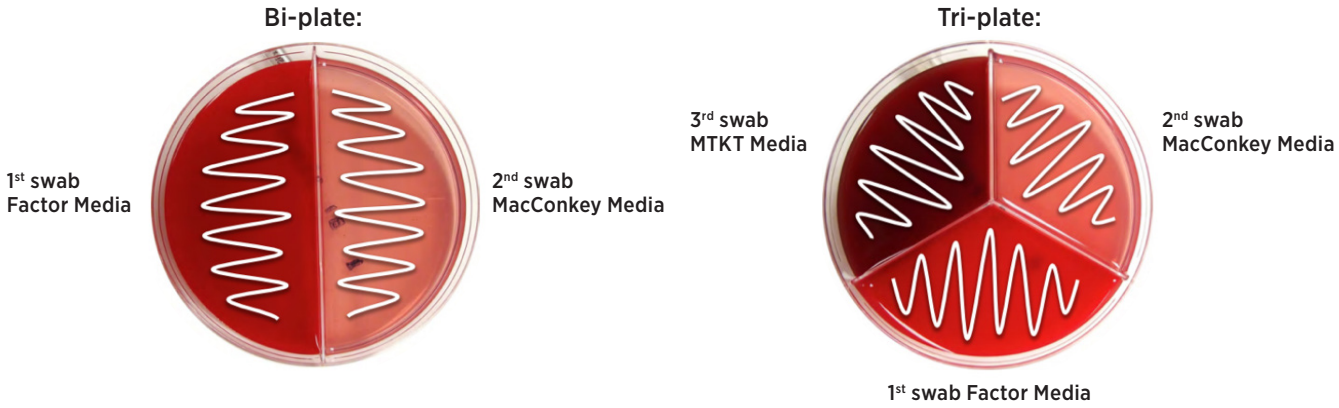

Figure 4. *Swab pattern for bi-plate (a) and tri-plate (b) (photo credit Minnesota Easy Culture System).*

10. Re-dip the cotton swab for each media section. For a bi-plate, dip the cotton swab once more or for a tri-plate dip it twice more. Repeat the side-to-side plating motion (Figure 3 and 4).

11. Once all sections are swabbed, place the lid back on the plate and replace the cap on the milk sample. Freeze the milk sample in case the media plate was contaminated during plating.

12. Place the plate in the incubator upside down (the lid side down with the agar facing down, see Figure 5) and check for initial growth at 24 hours. Use the Minnesota Easy Culture System User's Guide to identify any bacteria present and record the results. Place the plate back in the incubator.

13. After 48 hours, check for any new bacteria growth and use the Minnesota Easy Culture System User's Guide to identify any new bacteria present. Record all results in your record book.

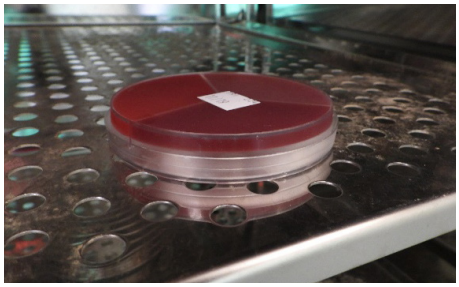

Figure 5. *Place the plate in the incubator lid side down (photo credit Kody Hash).*

For more information on starting on-farm culturing on your operation, contact your local county Extension agent or Liz Eckelkamp at eeckelka@utk.edu or 865-974-8167.

Resources

University of Minnesota Veterinary Diagnostic Laboratory. 2013. Minnesota Easy Culture System User's Guide. https://www.vdl.umn.edu/sites/vdl.umn.edu/files/mn-easy-culture-system-ii-users-guide-english.pdf

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