Testing Bulk Tank Milk Samples
G. M. Jones and Susan Sumner*

Samples of bulk tank milk are collected regularly and milk quality tests are performed by milk coops, plants, or Virginia Department of Agriculture and Consumer Services. Some milk coops offer bulk tank profiles which involve several tests related to milk quality. The various tests are briefly described below, as well as a list of goals for high quality milk and conditions which adversely affect test results. Hopefully, an explanation of the use of these tests will help Virginia dairy farmers produce high quality milk, which has been defined by the Extension Milk Quality Leadership Council as milk with:

- Somatic cell count equal to or less than 200,000
- Standard plate count equal to or less than 5,000
- Preliminary incubation count equal to or less than 10,000
- Drug residue free
- Sediment count equal to or less than 1.0
- Cryoscope reading equal to or less than -.530° H

**Standard Plate Count (SPC)** determines the number of visible colony-forming units (numbers of individual or tightly associated clumps of bacteria) in 1 ml. of milk incubated at 90° F for 48 hours. If SPC exceeds 5,000 cfu/ml., there is usually a reason. SPC is usually less than 5,000 cfu/ml. if sanitation is good and cooling is adequate. A SPC of 10,000 should be achievable by most farms (Murphy, 1997). The Extension Milk Quality Leadership Council has recommended that quality milk should have a SPC of 5,000 and less. In a test of 10,662 Virginia bulk tank samples by Virginia Department of Agriculture and Consumer Services (VDACS) between December 1, 1997, and November 16, 1998, 59% were below 5,000, 76% were below 10,000, and 87% were below 20,000. In New York State, of 855 bulk tank samples, 30% were below 5,400 cfu/ml and 50% were below 10,000 (Boor et al., 1998). The regulatory accepted limit is 100,000, although industry standards are often 50,000. Most high raw milk bacteria counts involve improper cleaning and sanitizing of dairy equipment, poor production practices (especially dirty cows), inadequate cooling, mastitis problems, or improper handling of sample by hauler or carrier of sample from plant to lab.

**Preliminary Incubation Count (PICount)** - A milk sample is incubated at 55° F for 18 hours, followed by the SPC procedure. PICount is considered the best measure of raw milk keeping quality and sanitation practices on farms. These organisms are usually associated with off-flavors, milk spoiling, and short shelf-life (Dairy Practices Council). PICount should not exceed 100,000 cfu/ml. Desirable PICount is less than 25,000 cfu/ml. The Extension Milk Quality Leadership Council has recommended that quality milk should have a PICount of 10,000 and less. Of 855 bulk tanks tested in New York, only 30% were less than 26,000 and 10% were below 8,000 cfu/ml. VDACS does
not conduct PICounts but several milk coops do and use them in their quality premium payment programs.

Listed below are possible causes of high PICounts. These also could apply to high SPC.

- Failure to thoroughly clean equipment after each use or neglecting to sanitize equipment before using (a major cause)
- Slow cooling farm bulk tank or temperature above 40°F (bulk tanks should be less than 40°F within two hours of milking and kept below 45°F during subsequent milkings). This points out the importance of the recording thermometer and its use.
- Problems with debris buildup in plate coolers and chillers.
- When milking fresh cows and problem cows in bucket milkers, hoses need to be kept clean
- Dirty animals; may need clipping
- Poor udder sanitation practices (dirty, excessive water used to wash teats & udders); teats need to be clean, sanitized, and dry
- Contaminated water supply, especially coliforms or other spoilage-causing bacteria
- Wash water temperature should start at 155-170°F and drain at above 120°F
- Gaskets and rubber parts need to be clean, free of cracks and deposits, and replaced when necessary
- Improperly drained milking equipment
- Teatcup liners should be clean and free of cracks and changed on schedule
- Pulsator and main vacuum supply lines need cleaning on a regular basis and especially whenever milk enters the line
- *Streptococcus agalactiae* or environmental streptococcus mastitis infections

**Lab Pasteurized Count (LPC)** measures the number of bacteria that survive lab pasteurization at 145°F for 30 minutes. Levels in raw milk should be less than 250-300 cfu/ml. Bacteria may be Bacillus species; usually it does not include mastitis-causing bacteria. Of 855 bulk tank samples analyzed in New York State, 60% were below 200 and 70% were less than 300 cfu/ml. These thermotolerant (heat loving) bacteria may cause spoilage in pasteurized milk. High LPC can be caused by unclean equipment and/or improper sanitizing practices, as well as leaky pumps, old pipeline gaskets, inflations and other rubber parts, and milkstone deposits (Murphy, 1997). VDACS does not perform this test. Some milk coops include LPC as a part of the bulk tank profile.

**Coliform Count** provides an indication of unsanitary production practices and/or mastitis infection. A count of less than 100 cfu/ml. is considered acceptable. Counts of 10 cfu/ml. or less are achievable and desirable. Of the NY bulk tank samples, 10% were less than 10 and 70% were below 66 cfu/ml. Udders and teats may be dirty and premilking hygiene may be inadequate. This test is not a part of VDACS’ regulatory testing program.

Contamination from the exterior of the udder can influence LPCs, PICounts, and coliform counts (Murphy, 1997). The degree of cleanliness of the milking system probably influences the total bulk milk bacteria as much as, if not more, than any other factor. Milk residue left on equipment contact surfaces supports the growth of a variety of microorganisms. Environmental contaminants (i.e. from bedding, manure, feeds) are more likely to grow on soiled equipment surfaces. Less efficient cleaning, using lower temperatures and/or the absence of sanitizers tends to select for the faster growing, less resistant organisms, principally Gram-negative rods (coliforms and Pseudomonads) and lactic streptococci. This will result in high PICounts and in some cases elevated LPCs.
Cryoscope or Freezing Point- Over -.530° Horvet suggests milk contains some added water or composition has been altered somehow. In New York, less than 3% of bulk tanks had freezing points greater than -.530. From December, 1997, through May, 1998, VDACS found only 16 violative samples in 5,631 tests or 0.28%.

Possible causes of high cryoscope readings include:
- Intentional or accidental addition of water
  - Chasing milk from receiver jar
  - Poor milking system drainage
  - “Wet cow” milking or use of excessive water
  - Backflushing units with vacuum on
  - Bulk tank valve should be open to drain
  - Rinsing top of tank
- Faulty gaskets in plate coolers (some brands)
- Freezing of milk in bulk tank
- Thin or under-conditioned cows because of grazing or low concentrate or energy intakes

Sediment- Acceptable levels are less than 1.5 mg./gal. milk. Cows’ udders should be clipped, teats and udders need to be clean, and teatcups properly attached and removed. Large herds need to stop during milking and change filters. Although this test isn’t conducted by VDACS nor by all milk coops, the Extension Milk Quality Leadership Council recommends that high quality milk should contain 1.0 or less.

Rancidity causes objectionable flavor with sharp, unclean, astringent taste that lingers as an unpleasant aftertaste. Psychrotrophic organisms such as Pseudomonas, Aerobacter, and Bacillus are harbored in crevices of pipelines, gaskets, etc., as a result of poor cleaning and incomplete sanitation, and they produce lipases which may not be destroyed by pasteurization. Lipases hydrolyze (break down) fats to glycerol and free fatty acids. It is expressed as acid degree value (ADV) which should not exceed 1.0. This test is conducted by some processors, but it’s not done by all milk coops.

Possible causes of highADV:
- Violent agitation, such as excessive pumping, or excessive agitation caused by slow cooling in farm bulk tank, pipeline obstructions, or freezing in bulk tank The elimination of risers in milk lines and the use of vacuum tanks have removed some of this problem.
- Airleaks in pipelines
- Leaky valves
- Flooded milklines, high milklines
- Teatcup liner slippage or excessive air admission
- Late stage of lactation weakens fat globule membrane’s protective lecithin-protein layer

Milk Cultures- Culturing of bulk tank milk samples can be useful to determine the presence or absence of certain bacteria and may be conducted by milk plants, VDACS, veterinary services, or the Virginia-Maryland Regional College of Veterinary Medicine. Changes in bulk tank cultures will monitor the impact of management changes upon milk quality and infection status. Samples taken over consecutive days or weeks give a clearer picture of a herd’s problem than a sample from one bulk tank of milk. Agitate the milk in the tank for 5 minutes prior to collection (National Mastitis Council or NMC). Use a clean, sanitized dipper to collect the milk sample from the top of the tank (never the outlet). Pour the first sample back into the tank to rinse sanitizer off the dipper. Sample two consecutive bulk tanks and freeze the samples. Sample the third bulk tank load and refrigerate it until all three samples can be delivered to a laboratory. The presence of Staphylococcus aureus or Streptococcus agalactiae almost always indicates the presence of these kind of mastitis infections in the herd.

Staphylococcus aureus is shed in low numbers by infected quarters and generally does not cause high bacteria counts (SPC) in bulk tank milk, but bulk tank somatic cell count (SCC) would be expected to be elevated. NMC states that it should be possible to attain a bulk tank Staphylococcus aureus count of less than 50 cfm/ml.
The presence of *Streptococcus agalactiae* in bulk tank milk is due only to infected udders. Both bacteria count (SPC) and SCC could be elevated by the presence of this infection in the herd. Eradication of this pathogen from the herd is possible.

Environmental streptococci and coliform (usually *E. coli*) counts above 1,000 and 500 cfu/ml, respectively, indicate poor hygiene either during equipment cleaning and sanitation, during milking, or between milkings. Common contaminants are bedding, manure, soil, or water. According to NMC, milking wet udders, organic soil buildup in milklines, cracked gaskets and inflations, inadequately heated wash water, inadequate cooling of milk, and udder infections all contribute to high environmental streptococcal or coliform counts in bulk milk. Udder infections should be highly correlated to cases of clinical mastitis in the herd. Bacteria counts (SPC) may be high while SCC are low.

Once results of bulk tank cultures are known, milk samples should be collected from individual cows and cultured. Take aseptic samples from cows with high DHI SCC or positive CMT, cows with clinical mastitis, and fresh cows, especially heifers. Samples can be stored in the freezer for as long as 6 weeks. Individual cow sampling can sometimes be disappointing because no growth can occur with milk from problem cows. According to the NMC (1987), 25-40 percent of all clinical samples can be negative on routine culturing. The reasons include:

- Numbers of certain organisms, such as mycoplasma, *Staphylococcus aureus*, and coliforms, can vary greatly in infected quarters, and may occasionally be less than the minimum detection limit of the assay. The minimum detection limit when plating 0.01 ml of milk is about 100 colony forming units per ml.
- The organism may no longer be present and the clinical signs are due to by-products such as endotoxins.
- Somatic cells may have phagocytized the organisms.
- Antibiotics may have killed or suppressed organism numbers to unrecoverable levels.
- Storage may have reduced numbers of viable organisms to undetectable levels.
- The organism may require cultural conditions other than those used for isolation (i.e. reduced temperature, prolonged incubation, special media, anaerobic conditions, etc.).

Three bulk tank samples were collected from a herd where the DHI SCC score averaged 3.36, with an actual SCC average of 537,000 and 28% of the cows had high SCC (score 5 and above or actual SCC above 284,000). The herd was treating 1-2 cows per week for clinical mastitis. At the previous month’s test, the average SCC score was 3.12, or actual SCC averaged 477,000 and 20% of cows had high SCC. This suggests that the herd’s subclinical mastitis is increasing. The bulk tank cultures had low coliform and streptococci counts accompanied by low bacteria counts. Two bulk tank samples had *Staphylococcus aureus* counts higher than desired. Staph infected herds often have high SCC but low bacteria counts. The dairy producer felt that the herd had symptoms characteristic of a herd with Staph infections. However, if bulk tank sample C had been the only sample collected, results would have been inconclusive and perhaps misleading.

<table>
<thead>
<tr>
<th>Bulk tank</th>
<th>Streptococci per ml</th>
<th>Coliform per ml</th>
<th><em>Staphylococcus aureus</em> per ml</th>
<th>Bacteria count per ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Desired</td>
<td>&lt; 1,000</td>
<td>&lt; 500</td>
<td>&lt; 50</td>
<td>&lt; 5,000</td>
</tr>
<tr>
<td>A</td>
<td>320</td>
<td>340</td>
<td>80</td>
<td>740</td>
</tr>
<tr>
<td>B</td>
<td>840</td>
<td>40</td>
<td>120</td>
<td>1,000</td>
</tr>
<tr>
<td>C</td>
<td>760</td>
<td>0</td>
<td>40</td>
<td>800</td>
</tr>
<tr>
<td>Type of Bacteria</td>
<td>Source</td>
<td>Comments</td>
<td>High SCC</td>
<td>High SPC</td>
</tr>
<tr>
<td>----------------------------------</td>
<td>---------------------------------------------</td>
<td>-----------------------------------------------</td>
<td>----------</td>
<td>----------</td>
</tr>
<tr>
<td><em>Staph. aureus</em> (coagulase positive)</td>
<td>Infected quarters, Teat lesions, new additions to herd</td>
<td><strong>Contagious</strong>: hands, wash rags, teatcup liners</td>
<td>**</td>
<td></td>
</tr>
<tr>
<td>Staph species (coagulase neg.)</td>
<td>Normal on teat skin, bedding, low SCC</td>
<td>Dry treat all cows, predip &amp; postdip with germicidal teat dip</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Str. agalactiae</em></td>
<td>Infected quarters, new additions to herd</td>
<td><strong>Contagious</strong>: hands, wash rags, teatcup liners</td>
<td>**</td>
<td>**</td>
</tr>
<tr>
<td>Environmental streptococci</td>
<td>Manure, bedding, wet &amp; dirty teats, liner slips</td>
<td>Wet &amp; dirty conditions that expose teat ends to bacteria; dirty housing and calving environments</td>
<td>*</td>
<td>**</td>
</tr>
<tr>
<td>Coliforms (<em>E. coli, Klebsiella</em>)</td>
<td>Manure, bedding, wet &amp; dirty teats</td>
<td>Clipped, clean, and dry udders &amp; teats; freestalls</td>
<td>*</td>
<td>*</td>
</tr>
<tr>
<td>Mycoplasma</td>
<td>Infected quarters, new additions to herd</td>
<td><strong>Contagious</strong>: hands, wash rags, teatcup liners</td>
<td>**</td>
<td></td>
</tr>
<tr>
<td>Pseudomonas</td>
<td>Water, soil, manure</td>
<td>Found in dirty milking equipment, contaminated water, contaminated antibiotics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prototheca</td>
<td>Water, manure, infected quarters</td>
<td>No treatment; eliminate stagnant water &amp; manure</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serratia</td>
<td>Contaminated water, bedding or litter</td>
<td>Eliminate washing of udders &amp; teats</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nocardia</td>
<td>Infected quarters</td>
<td>Found in contaminated treatments &amp; multi-dose</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yeast &amp; molds</td>
<td>Infected quarters</td>
<td>Found in contaminated treatments &amp; multi-dose</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
References:


National Mastitis Council. 1987. Laboratory and Field Handbook on Bovine Mastitis, Madison, WI.