

ELECTRONIC SOMATIC CELL COUNT

Fossomatic™ Minor (Raw Commingled Cow, Sheep, Goat, Water Buffalo and Camel Milk) IMS #16

(Unless otherwise stated all tolerances $\pm 5\%$)

1. **Laboratory Requirements (see Cultural Procedures (CP) items 33 & 34)** _____
 - a. Un-preserved samples may be tested up to 72 hours after initial collection _____
 - b. Samples may be tested up to 7 days after initial collection if preserved with 0.02% 2-bromo-2-nitropropane-1,3-diol (Bronopol™) or 0.05% potassium dichromate ($K_2Cr_2O_7$) _____

2. **Comparative Test with DMSCC**
[NOT required as a co-requisite for certification of analysts in laboratories purchasing standards from a CERTIFIED provider (item 9.b)] _____
 - a. Analyst(s) certified for DMSCC _____
 - b. Each analyst seeking certification for the ESCC test shall perform the comparative test _____
 1. Test 4 samples (100K-200K, 300K-500K, 600K-800K and 900K-1.2M) in triplicate for both DMSCC (three separate smears each) and ESCC _____
 2. Results must be evaluated by State/Federal LEO and shown to be acceptable prior to official use of test in laboratory _____
 3. Copy of comparison and results in QC record (or easily accessible on file in the laboratory); kept for as long as analyst is certified _____
 - c. Required for laboratories preparing in house standards or using commercially prepared standards (items 9.a and c) and for those testing goat or camel milk _____

APPARATUS

3. **See CP items 1-4** _____
4. **Water Bath** _____
 - a. Circulating and thermostatically controlled to 37-42°C _____

REAGENTS

5. Reagents

- a. Dye Solution Lot #: _____ Exp. Date: _____
- b. Clean 1 Lot #: _____ Exp. Date: _____
- c. Clean 2 Lot #: _____ Exp. Date: _____

6. Preparation

- a. Ready to Use Dye Solution: Pour into a clean glass container designated for the Dye Solution (item 5.a). Use within 4 weeks of dispensing into container.

Date Dispensed: _____ Exp. Date: _____

- b. Clean 1 Solution: In a clean glass container, mix one unit (20 mL) Clean 1 (item 5.b) with deionized (DI) or Microbiologically Suitable (MS) water to make 1 L, store and use within 4 weeks; when stored at 2-8°C, use within 8 weeks

Date Prep.: _____ Exp. Date: _____

- c. Ready to Use Clean 2 Solution: Pour into a clean glass container designated for the Clean 2 Solution (item 5.c). Use within 4 weeks of dispensing into container.

Date Dispensed: _____ Exp. Date: _____

7. All solutions labeled with date prepared and expiration date

START UP

8. Cell Counter

- a. Check that the volume of Dye, Clean 1 and Clean 2 solutions in the supply containers is sufficient for the number of samples to be tested
- b. Solutions not used beyond expiration date(s)
- c. Perform the "Start Up" Job sequence: If the Zero Count is > 6, repeat "Clean Cuvette" and re-check the zero
- d. **IF ANY ABOVE PARAMETERS ARE OUT OF VARIANCE, CORRECT BEFORE PROCEEDING**
- e. Maintain records on all parameters each time instrument is used

9. Milk Standards

a. Commercially prepared: _____

Lot#: _____ Date Rcd: _____

1. Four standards in ranges 100K-200K, 300K-500K, 600K-800K and 900K-1.2M
2. Perform DMSCC in triplicate on each standard in set and average counts; maintain records
3. Perform DMSCC check in rotation by all certified analysts
4. Use standards within one week

Lab Exp. Date: _____

b. Certified provider: _____

Lot#: _____ Exp. Date: _____ Date Rcd: _____

1. Four standards in ranges 100K-200K, 300K-500K, 600K-800K and 900K-1.2M
2. Maintain copies of all provided DMSCC values
3. Measure and maintain records of temperature (0.0-7.5°C) of standards as received
4. Maintain copies of all correspondence regarding problems
5. Standards used by manufacturer's expiration date
6. Failed standards shall be verified with DMSCC

a. If no analysts certified for DMSCC then a new set of standards is required

b. Do not continue with official testing until the new standard(s) test(s) in range

c. Laboratory prepared (weekly)

1. Prepare from raw milk > 18 hours old preserved with 0.05% potassium dichromate ($K_2Cr_2O_7$)
2. Or, preserved with 0.02% 2-bromo-2-nitropropane-1,3-diol (Bronopol™)
3. Standards **cannot** be preserved with formalin

4. Prepare 4 standards in ranges 100K-200K, 300K-500K, 600K-800K and 900K-1.2M; use within one week

Lab Prep. Date: _____ Lab Exp. Date: _____

5. Perform DMSCC in triplicate on each standard and average counts; maintain records

6. Perform DMSCC check in rotation by all certified analysts

d. Hourly Control Sample (instrument drift check)

1. Use one of the standards (items 9.a, b or c) in the 600-800K range, test in triplicate and determine average

2. Optionally, prepare sufficient control/sample 600-800K range, test in triplicate and determine average

PROCEDURE

10. Testing Standards (each time instrument used)

- a. Heat standards to 37-42°C (using a temperature control) and test within 30 min of reaching temperature, use once and then discard, i.e. do not re-use

- b. Mix by inverting at least 10x, test standards within 3 min

- c. Test the standards in triplicate and average the counts for each level; maintain records

- d. Each standard's average must be within 10% of the DMSCC (item 9) for that level, except within 15% for 100K-200K standard; maintain records

- e. Repeatability - a standard in the 300K to 800K range must have a coefficient of variation (CV) of 5% or less on 10 replicates (**Refer to Operating Manual**); maintain records

- f. Alternatively, set and run standard check as a "Custom Job", enter DMSCC values (item 9) into Excel™ macro, starting the job will enable 10.c through 10.e to be run and calculated automatically

- g. **THESE PARAMETERS MUST BE ACHIEVED BEFORE PROCEEDING**

11. Testing Samples

- a. Heat samples to 37-42°C (using a temperature control) and test within 30 min of reaching temperature

- b. Test samples within 10 min after removal from water bath

- c. Mix by inverting at least 10x, test samples within 3 min

d. Record number of cells counted for each sample _____

12. With Continuous Operation: _____

a. Perform a zero check (item 8.d) hourly _____

b. Test a standard or optionally a control/sample (item 9) in the 600K to 800K range hourly in triplicate and determine the average, must be within 5% of the original established instrument average value (optionally, within 10% of original DMSCC average) _____

c. Maintain records _____

13. Routine maintenance _____

a. Maintain records _____

REPORTING

14. Computing and Reporting Counts _____

a. Count obtained x 1000 is the cell count/mL milk _____

b. In reporting electronic somatic cell counts (ESCC/mL); record only first two left hand digits, raising second digit to next higher number when third digit is 6 or more _____

c. Report the two left hand digits (rounded) _____

1. If the third digit is 5 the second digit is rounded by the following rule _____

a. When the second digit is odd round up, raise the second digit by 1 (odd up, 235 to 240) _____

b. When the second digit is even round down, delete the 5 and report the second digit as is (even down, 225 to 220) _____

d. If count on instrument is < 100 report as < 100,000 ESCC/mL _____

e. If goat or camel milk is over the regulatory limit, follow confirmation procedure in PMO _____