

**APPENDIX N BULK MILK TANKER SCREENING TEST FORM**

**NEOGEN BETASTAR ADVANCED FOR BETA-LACTAMS TEST  
(Raw Comingled Cow Milk)  
IMS #9-N3**

**[Unless otherwise stated all tolerances are  $\pm 5\%$ ]**

**GENERAL REQUIREMENTS**

1. See Appendix N General Requirements (App. N GR) items 1-8 & 15 \_\_\_\_\_

**SAMPLES**

2. See App. N GR item 9 \_\_\_\_\_

**APPARATUS & REAGENTS**

**3. Equipment** \_\_\_\_\_

- a. Neogen Corporation Raptor© Integrated Analysis Platform (Manual available).  
Thermostatically controlled at  $65.0 \pm 5.0^\circ\text{C}$  \_\_\_\_\_

Serial Number: \_\_\_\_\_

1. Temperature checked daily on the screen and printout (day of use),  
Records maintained (Printout acceptable for daily temperature check) \_\_\_\_\_

a. Incubator Temperature: \_\_\_\_\_

b. Annual temperature verification performed; records maintained \_\_\_\_\_

1. Date of last verification: \_\_\_\_\_

- b. Reader calibrators \_\_\_\_\_

1. Positive: \_\_\_\_\_

2. Negative: \_\_\_\_\_

- c. Pipettor – 400  $\mu\text{L}$  and disposable tips (see App. N GR item 7) \_\_\_\_\_

1. **FOR SCREENING ONLY** - Disposable 400  $\mu\text{L}$  single-use poly-pipets \_\_\_\_\_

**4. Test Kits** \_\_\_\_\_

- a. BetaStar Advanced Test for Beta-lactams Kit \_\_\_\_\_

Lot #: \_\_\_\_\_ Exp. Date: \_\_\_\_\_

QC Date: \_\_\_\_\_ By: \_\_\_\_\_

## 5. Sample and control agitation

- a. Mix milk sample(s)/control(s) 25 times in 7 sec with a 1 ft movement or vortex for 10 sec at maximum setting; use within 3 min (samples/controls must be in appropriate containers to allow the use of vortexing)

## 6. Reagent Stability and Preparation

- a. Test Kit including strips are received under ambient temperature
- b. Strips stored at 18 - 30°C (64 - 86°F), maintain no longer than manufacturer's expiration date
- c. Negative Control
1. Previously negative tested raw milk
  2. Milk can be screened (previously tested) by the testing location making and/or using the controls
  3. Negative control must result in a ratio of  $\geq 1.15$  for both the beta-lactam and ceftiofur test lines; maintain records

Sample ID: \_\_\_\_\_ Date Tested: \_\_\_\_\_

Record test line values (Ratio): \_\_\_\_\_

Beta-lactam line: \_\_\_\_\_

Ceftiofur line: \_\_\_\_\_

4. Use within 72 hours when maintained at 0.0-4.5°C
5. Or, aliquot within 24 hours and freeze at -15°C or colder in a non-frost-free freezer or in an insulated foam container in a frost-free freezer; use within 2 months

Lab Prep. Date: \_\_\_\_\_ Lab Exp. Date: \_\_\_\_\_

- a. Thaw slowly in refrigerator or more rapidly in cold water. Mix well until sample is homogeneous

1. **Do Not use if there is visible protein precipitation**

- b. Store at 0.0-4.5°C and use within 48 hours. Do not refreeze

6. Day of use must result in a ratio of  $\geq 1.15$ ; maintain records

**Do Not proceed if out of range**

d. Positive Control - Manufacturer supplied, maintain no longer than manufacturer's expiration date

1. Lyophilized  $5.0 \pm 0.5$  ppb Penicillin G /  $100 \pm 10$  ppb Desfuroyl ceftiofur

Lot #: \_\_\_\_\_ Exp. Date: \_\_\_\_\_

2. Store according to label instructions

3. Reconstitute with 1.0 mL of fresh or previously frozen previously screened beta lactam negative raw commingled cow milk

4. Positive control must produce a ratio of  $\leq 0.85$  for both the beta-lactam and ceftiofur test lines; maintain records

Record test line values (Ratio):

Beta-lactam line: \_\_\_\_\_

Ceftiofur line: \_\_\_\_\_

5. Store reconstituted positive control at 0.0-4.5°C for no more than 48 hours

6. Or, aliquot within 24 hours and freeze at -15°C or colder in a non-frost-free freezer or in an insulated foam container in a frost-free freezer; use within 2 months. **Do Not** freeze positive control if it was made with previously frozen negative control

Lab Prep. Date: \_\_\_\_\_ Lab Exp. Date: \_\_\_\_\_

a. Thaw slowly in refrigerator or more rapidly in cold water. Mix well until sample is homogeneous

1. **Do Not use if there is visible protein precipitation**

b. Store at 0.0-4.5°C and use within 24 hours; do not refreeze

7. Day of use must produce a ratio of  $\leq 0.85$ ; maintain records

**Do Not proceed if out of range**

## TECHNIQUE

### 7. Daily Performance and Operation Check

a. See App. N GR items 10.b-d

b. Raptor® Integrated Analysis Platform

1. At Raptor® start-up, calibration of camera and LED occurs automatically when instrument is turned on

2. If the calibration is unsuccessful, the reader will not operate. A warning message will prompt the user, "Calibration unsuccessful. Contact Neogen" \_\_\_\_\_
3. Annual calibration defines x and y offsets for the Raptor system \_\_\_\_\_
  - a. User performed annual calibration is required every 365 days. Verify annual calibration was performed within last 365 days. Please see user manual for more details \_\_\_\_\_  
Date of last calibration: \_\_\_\_\_
4. Daily reader check calibration \_\_\_\_\_
  - a. The reader check calibration must be performed daily in each of three ports in the Raptor System \_\_\_\_\_
  - b. There are three calibration test strips within each cartridge, all positive or all negative \_\_\_\_\_
  - c. Both positive and negative calibration cartridges must read within the limits specified  $\leq 0.85$  for positive and  $\geq 1.15$  for negative; maintain records \_\_\_\_\_
  - d. Positive Calibrator Ratios: (Specification  $\leq 0.85$ ) \_\_\_\_\_  
Port 1: \_\_\_\_\_ Port 2: \_\_\_\_\_ Port 3: \_\_\_\_\_
  - e. Negative Calibrator Ratios: (Specification  $\geq 1.15$ ) \_\_\_\_\_  
Port 1: \_\_\_\_\_ Port 2: \_\_\_\_\_ Port 3: \_\_\_\_\_
5. If reader check calibrations are out of range, contact Neogen before proceeding \_\_\_\_\_

## 8. Test Procedure

- a. Make sure hands are clean and dry before handling test kits \_\_\_\_\_
- b. Set out required number of cartridges and place them in a dry labeled container at room temperature, or take out cartridges as needed \_\_\_\_\_
  1. Cartridges that have been removed from the protective storage container must be kept clean and dry \_\_\_\_\_
  2. Any cartridges removed from the kit that remain unused at the end of the testing day must be discarded \_\_\_\_\_
- c. Cartridges are pre-loaded with one test strip. Up to two more test strips for other residues may be loaded into the cartridge. One cartridge, loaded with up to three test strips, can be used to test one milk sample \_\_\_\_\_

- d. Place cartridge with test strip(s) into any of the three ports. When cartridge is inserted into the port, the port will automatically begin to adjust to the proper temperature \_\_\_\_\_
- e. The bar code on the test device will be read. If the QR (quick response) code for the lot of strips has not been entered into the system, the bar code reader in the front of the reader will turn on automatically. Scan the QR code found on the container storing the test strips \_\_\_\_\_
- f. Instrument will prompt user for the milk sample ID. Scan or enter the sample ID at this time \_\_\_\_\_
- g. Mix milk sample(s)/control(s) (See item 5.a) \_\_\_\_\_
- h. The user will be prompted to add the milk sample when the port reaches  $65.0 \pm 5.0^{\circ}\text{C}$ . **Do Not** add milk sample until prompted to do so \_\_\_\_\_
- i. Add 400 uL of mixed sample/control into the back of the cartridge \_\_\_\_\_
  - 1. Using pipettor (item 3.c) with a new tip for each sample/control and holding pipettor vertically draw up 400  $\mu\text{L}$  avoiding foam and bubbles \_\_\_\_\_
    - a. Remove tip from liquid \_\_\_\_\_
    - b. While holding the pipettor vertically, expel test portion into cartridge \_\_\_\_\_
    - c. After sample is delivered into cartridge, eject pipettor tip into the back of the cartridge to prevent double loading of the same sample or loading a second sample into the same cartridge \_\_\_\_\_
  - 2. **FOR SCREENING ONLY** - Using a new manufacturer provided single-use 400  $\mu\text{L}$  poly-pipet (item 3.c.1) for each sample/control \_\_\_\_\_
    - a. Squeeze top bulb while holding single-use pipet vertically and draw up test portion avoiding foam and bubbles. Insure that pipet shaft is completely full and sample overflows into the bottom half of the overflow reservoir \_\_\_\_\_
    - b. Remove tip from liquid \_\_\_\_\_
    - c. While holding the single-use pipet vertically, expel test portion slowly into the back of the cartridge. Excess portion should remain in reservoir \_\_\_\_\_
    - d. After loading milk sample into the cartridge, leave the used pipet in the back of the cartridge. This will prevent double loading the same sample or loading a second sample into the same cartridge \_\_\_\_\_
- j. Press “Next” after sample has been added. The unit will begin the 5 minute incubation after the system identifies the fluid front of the sample wicking up the device \_\_\_\_\_

- k. After 5 minutes the result will be displayed on the screen, an audible tone will sound, and the test result will automatically print \_\_\_\_\_
- l. Remove cartridge containing test strip(s) from the reader and discard the entire cartridge \_\_\_\_\_

**9. Interpretation with Reader** \_\_\_\_\_

- a. If there is a ratio of  $\geq 1.00$  on the reader, sample is **a Negative (NF)** \_\_\_\_\_
- b. If there is a ratio of  $< 1.00$  on the reader, sample is **an Initial Positive** \_\_\_\_\_

**10. Verification of Initial Positive Tanker Samples (see App. N GR item 11)** \_\_\_\_\_

**11. Confirmation of Presumptive Positive Tanker Samples (see App. N GR item 12) [Only in an accredited laboratory or by a CIS]** \_\_\_\_\_

- a. For Beta-lactam confirmation, run tests using one Beta-lactam strip per Cartridge \_\_\_\_\_

**12. Traceback of Producer(s) on a Confirmed Positive Tanker (see App. N GR item 13) [Only in an accredited laboratory or by a CIS (refer to M-a-85 current revision for a listing of test kits to assure equivalence)]** \_\_\_\_\_

**13. Re-instatement of Producer(s) [Only in an accredited laboratory or by a CIS (refer to M-a-85 current revision for a listing of test kits to assure equivalence)]** \_\_\_\_\_

**14. Reporting (see App. N GR item 14)** \_\_\_\_\_