

SOP #: 307.02Title: **SOP -** Quality Testing of Water

Approvals:

Attending Veterinarian

Date: 03/15/2018

### 1. Purpose

1.1 To describe the proper method for testing for the presence of bacteria in water utilizing two distinct methods:

1.1.1 Heterotrophic plate count or "HPC test

1.1.2 ATP Hygiene Monitoring System - SystemSURE Plus with Aquasnap swabs from the various watering systems in the Florida International University Animal Care Facilities.

1.2 Risks from bacteria detected in water

1.2.1 A number of studies have yielded virtually the same characteristic spectrum of heterotrophic bacterial strains. The predominant species in this spectrum are *Acinetobacter* spp., *Aeromonas* spp., *Alcaligenes* spp., *Comamonas* spp., *Enterobacter* spp., *Flavobacterium* spp., *Klebsiella* spp., *Moraxella* spp., *Pseudomonas* spp., *Sphingomonas* spp., *Stenotrophomonas* spp., atypical *Mycobacterium* spp., *Bacillus* spp. and *Nocardia*

### 2. Responsibility

2.1 It is the responsibility of ACF personnel to follow this procedure and supervisors for employee training and compliance.

### 3. Definitions

3.1 ACF - Animal Care Facility

3.2 RLU – Relative light units

### 3.3 CFU – Colony Forming Units

## 4. Guidelines

### 4.1 Heterotrophic plate count

- 4.1.1 Due to the transition to hydropacs to provide water to rodents, the heterotrophic plate count or “HPC test” is the preferred method of water testing.
- 4.1.2 Heterotrophs are broadly defined as microorganisms that require organic carbon for growth. They include bacteria, yeasts and molds. A variety of simple culture-based tests that are intended to recover a wide range of microorganisms from water are collectively referred to as “heterotrophic plate count” or “HPC test” procedures.
- 4.1.3 Microorganisms will normally grow in water and on surfaces in contact with water as biofilms. Growth following drinking-water treatment is normally referred to as “regrowth.” Growth is typically reflected in higher HPC values measured in water samples.
- 4.1.4 Elevated HPC levels occur especially in stagnant parts of piped distribution systems, in the plumbing system, and in plumbed-in devices, such as softeners, and carbon filters.
- 4.1.5 HPC measurements can be used to monitor the performance of filtration or disinfection processes.
- 4.1.6 Water analysis shall be performed on every hydropacs lot or more often if indicated by the Attending Veterinarian.
- 4.1.7 Testing procedure using HPC method
  - 4.1.7.1 Open the sampler package without touching the gridded filter surface. Write on sampler case with permanent marker the date and lot or location.
  - 4.1.7.2 Pour sample liquid into the sampler case filling to the upper 18 ml graduation.
  - 4.1.7.3 Insert the sampler paddle in the sample case for 30 seconds. If the entire membrane is properly wetted, the filter will appear dark gray. If immersed more than 30 sec, a loss of medium through the membrane and into the sample may occur.
  - 4.1.7.4 Remove the paddle and shake off the excess fluid. Empty the case and reinsert the paddle to form an airtight seal to prevent drying during incubation.
  - 4.1.7.5 Incubate sampler with gridded face down for 48-72 hours at 25-35°C
  - 4.1.7.6 After incubation is complete, remove paddle from case and examine filter surface with either a microscope or a magnifier.

- 4.1.7.7 Most colonies are glistening and translucent or transparent. Colors varies from colorless to white, cream, yellow or red.
- 4.1.7.8 An ideal count should be zero CFU.
- 4.1.7.9 To properly sample different portions of each water load a sample hydropac is taken at the start, the middle and the end of the pouch filling operation.  
Example: In a pouch collection of 600 pouches the technician should collect a pouch marked #1 from the beginning of the batch, pouch #2 at 300 pouches and pouch #3 at 600 for testing.
- 4.1.7.10 Each LOT is placed on hold until the QC testing is completed.
- 4.1.7.11 Disposal of sHPC sampler - can be disposed of in normal lab trash receptacles.

#### 4.2 ATP Hygiene Monitoring System - SystemSURE Plus with Aquasnap swabs

- 4.2.1 ATP Hygiene Monitoring System - SystemSURE Plus with Aquasnap swabs offers a sensitive and fast method of water quality control programs by detecting total ATP in water in seconds.
- 4.2.2 ATP presence in water is an indicator of microorganisms or organic material, which can have a direct impact on the health of the laboratory research animals. These tests are important in the assessment and maintaining quality of the water provided to the laboratory animals housed in the ACF.
- 4.2.3 Testing procedure using ATP SystemSURE Plus with Aquasnap
  - 4.2.3.1 Run tap water for 2-4 minutes.
  - 4.2.3.2 Remove one Aquasnap swab for each sample to be tested from foil bag.
  - 4.2.3.3 Allow swabs to come to room temperature before use. Do not remove cover from swab until it is ready for use.
  - 4.2.3.4 Aquasnap swabs are to be stored in the refrigerator at 2-8°C when not in use.
  - 4.2.3.5 Label swab with permanent marker indicating the following information:
    - 4.2.3.5.1 Location, room and/or collection source, date and time.
  - 4.2.3.6 Remove Aquasnap swab from the outer tube. Dip the swab for 5 to 10 seconds into a 20 mL sample of the water being tested. If the water is not homogenous or contains sediment, mix it thoroughly before sampling.

4.2.3.6.1 NOTE: Ultrasnap swabs used for environmental monitoring are designed primarily for surfaces but can be used for some types of water samples.

4.2.3.7 Reinsert swab into the swab tube. The device is now ready to be activated or can be left inactive for up to 4 hours in this state.

4.2.3.8 To activate, break the plastic valve at the top of the device by bending backward and forward. Squeeze the bulb twice to expel the liquid in the bulb to the bottom of the tube.

4.2.3.9 Bathe the swab bud in the liquid by shaking gently in a side-to-side motion for 5 seconds.

4.2.3.10 Place the entire swab device into the SystemSURE Plus luminometers and close the lid. This step must be done within one minute of activation.

4.2.3.11 Holding the luminometer in a vertical position, press 'OK' to initiate reading. The test result will appear on the screen in 15 seconds.

#### 4.2.4 Interpretation of Results.

4.2.4.1 The acceptable range (result) for water is 0 RLU.

4.2.4.2 If any reading above 0 (zero) is obtained, obtain a new sample and retest following the steps outlined previously.

4.2.4.3 If another unacceptable result is obtained inform the Attending Veterinarian. Positives can then be confirmed by mHPC (membrane Heterotrophic Plate Count).

4.2.4.4 Disposal of swabs – swabs are non-hazardous and can be disposed of in normal lab trash receptacles.

#### 4.3 Recording of Results.

4.3.1.1.1 Record the results on - Environmental Monitoring of Equipment, Surfaces and Water Quality - Form 926 current revision.

### 5. References

5.1 Hygiena ATP Monitoring Systems <http://www.hygiena.net/aquasnap.html>

5.2 [https://www.who.int/water\\_sanitation\\_health/dwg/HPCFull.pdf](https://www.who.int/water_sanitation_health/dwg/HPCFull.pdf)

### 6. Revisions

6.1 Rev. 02 – Updated the information on Heterophilic plate count method based on World Health Organization guidelines – March 2018