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An overview of methods for measuring the impact of sanitation procedures for swine transport vehicles

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Introduction: In today's swine industry, biosecurity protocols are perceived to be extremely important in order to protect the health status of a herd. While the easiest way to introduce a microorganism into a swine herd is through the introduction of infected animals, the potential for pathogen entry via contaminated fomites, i.e. boots, transport vehicles, etc. has been debated. Therefore, an extensive amount of time, labor and money is invested in the washing and disinfecting of facilities and transport vehicles, particularly in breeding stock operations, in order to reduce the risk of delivering unwanted pathogens along with a shipment of pigs. Unfortunately, there are no published protocols that attempt to measure whether such practices actually impact the microbial population; however, commercial kits are available from the food processing industry. The purpose of this paper is to review two methods, the RODAC System and the Lightning System that are currently being used to measure the cleanliness of swine transport vehicles following sanitation procedures. The author has experience with both systems, and their strengths and weaknesses will be summarized at this time:

A. Principles

1. The RODAC System

The RODAC System is based on the culture and quantification of bacteria isolated from a test surface. RODAC plates consist of Lethen agar, and a 16 square grid is enscripted on the plate. Following sample collection, the plate is incubated for 48 hours at 37 degrees C. The total number of bacterial colonies are counted, divided by 16 and results are reported as mean colonies per unit (grid square). Acceptable levels are less than 2.0 colonies per unit, with higher readings deemed to be unacceptable.

2. The Lightning System

The Lightning System (IDEXX Laboratories, Atlanta GA) is based on the detection of residual Adenosine Triphosphate (ATP), a compound present in organic debris, feces, feed, bacteria, protozoa, internal and external parasites, molds, yeasts, and fungi. It is not possible to measure the presence of viruses using this system. Each test kit is a self-contained unit, consisting of a swab for sample collection, a wetting agent and the enzyme luciferase. Luciferase is derived from fireflies and is activated in the presence of ATP. A hand-held luminometer is used to determine the amount of fluorescence emitted from the swab and to calculate the level of residual ATP present on the tested surface. Readings are displayed as numerical values ranging from 0 to 7.5. and categorized into "Zones of Cleanliness". The measuring system is based on a logarithmic base 10 scale; therefore, a reading of 4.5 is 10 times greater than a reading of 3.5, and a hundred times greater than a reading of 2.5. Readings of 0 to 2.5 are determined to be in the "Clean Zone", readings of 2.5 to 3.0 in the "Caution Zone", while readings > 3.0 were considered to be in the "Dirty Zone". The entire process of swabbing, processing the sample and the luminometer calculation takes approximately 15 to 20 seconds.

B. Method of sample collection

General comments

A thorough visual inspection should always initiate the investigation PRIOR to the use of a test kit. Frequently, the level of contamination can be determined visually, thereby eliminating the need for further testing and reducing cost. Surfaces selected for testing should include floors, walls, ceilings, gates and ramps of trailers, as well as interior surfaces of cabs (floor mats, foot pedals, etc). Trailer

Continued on back

surfaces should be divided into thirds (front, middle, and rear). It is recommended to sample 2 to 3 points during each sampling period, including one "high risk" point such as the tailgate of the trailer, as well as areas typically known to be hard to clean, such as ceilings, gates, or the front third of the trailer floor.

1. The RODAC System

The surface of the RODAC plate is domed, and the agar surface is gently applied to a selected area of the test surface or a swab can be used.

2. The Lightning System

The swab is removed from the test kit, applied to the desired surface, rotated over a 1 square foot area, and re-inserted in the test tube, allowing contact with the wetting agent, swab surface and enzyme.

C. Strengths and weaknesses

The following conclusions are derived from the author's experiences.

1. The RODAC System

Advantages-The ability to culture specific microorganisms occasionally proves to be advantageous if the need to determine the genus and species of bacteria present on the tested surface, or to carry out antimicrobial resistance or disinfectant sensitivity tests, is required.

Disadvantages-The need for a 48 hour incubation period is a major inconvenience when shipments are taking place on a daily basis. Furthermore, in the hands of the author, parallel testing of the RODAC and Lightning revealed a lack of sensitivity on the part of the former assay. Finally, the cost of the RODAC was determined to be approximately 3 times as great as a single Lightning test.

2. The Lightning System

Advantages-The turn-around time for results of this method was exceptional, as was the cost per test. Results could be effectively entered into computerized spreadsheets (table 1) to identify trends regarding problem vehicles or personnel, or even graphed using a Statistical Process Control format. Data can be described and tested using statistical methods. The luminometer also has extensive memory and data can be downloaded to other locations for storage and analysis. Positive controls are included to allow for daily calibration of the unit.

Disadvantages-The initial cost to purchase the system was approximately \$3000. It is extremely sensitive, and the Zones of Cleanliness recommended by the manufacturer may need re-calibration to allow for its application to transport vehicles. This can be done by determining the "background" level of the luminometer through the reading of uninoculated swabs, or the sampling of known sterile surfaces, such as the interior of previously unopened syringe cases or blood collection tubes. These readings may register 2.0 or higher. Finally, supplies such as swabs, and positive controls need to be continually purchased, at \$2.60 and \$6.40 per respective unit.

Table 1: An example of a Lightning System database

Date	Vehicle #	Wash Personnel	Reading 1 Tailgate	Reading 2 Floor (first 1/3)
5/26/98	100	JS	2.9	3.1
5/26/98	105	AB	2.1	2.5
5/26/98	102	JS	3.5	4.2
5/26/98	110	BR	2.7	2.4
5/26/98	115	JS	3.7	3.4
5/26/98	103	AB	2.4	2.7
5/26/98	104	JS	2.2	4.6
5/26/98	108	BR	3.7	5.8
5/26/98	109	AB	2.3	4.1
5/26/98	101	JS	6.5	5.1

Summary: Vehicles 102, 115, 104, 108 109 and 101 were unsatisfactory
JS was responsible for 4/6 unsatisfactory vehicles



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