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TESTING IRRIGATION WATER FOR GENERIC E. COLI Multiple Tube Fermentation (SM 9221B, E, F) and Quantitray (SM 9223B)

PURPOSE: Water used for production and harvest operations may contaminate lettuce and leafy greens if water containing human pathogens comes in direct contact with the edible portions of lettuce/leafy greens. Contamination may also occur by means of water-to-soil followed by soil-to-lettuce/leafy greens contact. Irrigation methods may have varying potential to introduce human pathogens or promote human pathogen growth on lettuce and leafy greens.

It is not feasible to routinely test irrigation water for every possible disease-causing microorganism. Instead, water quality standards are based on the concept of "indicator" organisms. According to this concept, irrigation water is tested for organisms that are not necessarily the cause of disease but are associated with contaminated water and indicate the potential for disease transmission. Testing for generic *E. coli* is considered the best available indicator of a fecal contaminated water source.

Quantitative testing of generic *E. coli* in a water source helps establish the extent of fecal contamination; higher levels of fecal contamination are associated with increasing health risk. The action levels for generic *E. coli* described in the GAP metrics should not be interpreted as distinguishing "safe" or "unsafe" levels—they should be used as benchmarks for reducing potential sources of contamination of lettuce and leafy greens.

SAMPLE COLLECTION: Water samples taken for coliform bacteria testing must be collected and handled carefully in order to ensure that the sample taken truly represents the bacteriological quality of water in the system. The following procedures will help you in this regard:

- 1. Sterile containers provided by your laboratory must be used. Do not touch or otherwise contaminate the inside of the container, the inside of the cap, or the threads of the container (e.g. do not place cap on the ground or in your pocket). The container contains a chemical to neutralize chlorine; do not rinse the container.
- 2. The April 18, 2007 GAP metrics specifies sampling as close to the point-of-use as possible (e.g. overhead sprinkler, valve used for pesticides/fungicide preparation, etc.). If water is found to be above action levels at this location, then additional testing of the source water and the initiation of a sanitary survey are required.
- 3. When you have found a suitable sample site, adjust the flow to minimize splashing if possible. Let water run for two or three minutes. Carefully fill the container up to the 100ml mark. Immediately replace the cap (tightly) and label the sample with sample site code (if applicable), description of sample point, date and time of collection, and name of sample collector.
- 4. Refrigerate the sample and/or place the sample in a cooler with sufficient ice until it can be delivered to the laboratory. It is required that samples arrive at the laboratory below 10 °C if received more than 1 hour after collection.

5. Complete the laboratory form including mailing address, name of sample collector, identification code, description of sample point, date and time of collection, and test ordered (i.e. "Coliform"). Note: use of consistent identification codes will facilitate data queries and determination of running geometric means.

CARE OF SAMPLE

<u>SAMPLES MUST BE SUBMITTED DIRECTLY TO THE LABORATORY WITHIN 6 HOURS OF</u> <u>COLLECTION.</u>

<u>REMEMBER: SAMPLES MUST BE COOLED AFTER COLLECTION UNTIL RECEIVED AT THE</u> <u>LABORATORY</u> (e.g. iced cooler).

Health department offices can provide for cold storage/transport from point of receipt.

INTERPRETATION OF RESULTS

It is impractical to use a single action level for generic *E. coli* for water used in all agricultural applications. For instance, water that contacts edible portions of plants should likely have more stringent standards than water that does not contact edible portions of plants. The following use-specific standards have been created for three uses determined to be most critical to lettuce and leafy green food safety:

Application	Maximum level for any single sample	Maximum level for geometric mean (rolling average of 5 most recent samples)
Pre-harvest foliar applications (e.g. overhead sprinkler irrigation, pesticides/fungicide application, etc.)	235 MPN/100 mL	126 MPN/100 mL,
Pre-harvest non-foliar applications (e.g., furrow or drip irrigation, dust abatement water).	576 MPN/100 mL	126 MPN/100 mL,
Post-harvest direct contact applications (e.g. re-hydration, core in field, harvest equipment cleaning, bin cleaning, product cooling, product washing)	<2 MPN/100 mL	

When calculating the geometric mean, lab results reported as less than the detection limit must be handled in a special way; these results cannot be treated as "0 MPN" since there is no logarithm for zero. The common way to handle this is to assign a numeric value of "1" for a result reported as "<1" or "<2" (Note: If the detection limit is greater than 2, assign a numeric value that is one-half the detection limit). Using the following hypothetical results for a five week period <1, <2, 18.5, 2, <1, the average of the logarithm for these values would be (0+0+1.3+0.3+0); or 0.32. The geometric mean would be 2.09 (or $10^{0.32}$).

Reference: Western Growers Association. April 25, 2006. Commodity Specific Food Safety Guidelines for the Lettuce and Leafy Greens Supply Chain.