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# Microtracers F
# Testing for Cross Contamination in Medicated Feeds

Please also refer to Literature Items "A-1" Quality Assurance With Microtracers F, "A-2" Microtracer "Rotary Detector", "A-3" Microtracers F- Quantitative Assays, and "C-9" The Use of Microtracers F in Coding the Presence of Coccidiostats in Poultry Feeds: Practical Implications.

**Preface:**

Medicated animal, poultry and aquatic feeds are often manufactured at feedmills making feeds for many species or if not at least many formulations for one species. Contamination of medications to non-target feeds is inevitable but quantifiable and controllable. Medicated feed assays, unfortunately, often do not offer a viable control mechanism because: they are expensive, they cannot be performed immediately and they are often inaccurate at contamination levels (i.e. 1 % or less of formulated levels).

Microtracers F (colored uniformly sized iron particles) offer a vehicle for locating and quantifying "cross contamination" of medicated feeds. Instead of running expensive and inaccurate drug assays, one uses one or more Microtracers F as indicators for the coded medication. The tracer is first mixed into the medicated feed premix, then the premix is added to the feed and mixed. Many samples are taken either only at truck loading or at many locations, depending upon the scope of the study. It is critical that feeds not formulated with the Microtracer follow the identical route at the feedmill as the "target" batch containing the Microtracer. Samples yielding significant levels of the tracer may then be analyzed for the medication to confirm (or refute) the preliminary results provided by the tracer.

Addition of the Tracer: The Microtracer is generally added to the medicated premix to yield 50 grams of tracer per metric tonne of final feed. Since the tracer has an expected count of 25,000 particles/gram, this means 50 x 25,000 or 1,250,000 colored iron particles are added to a one metric ton batch with 1,250 particles expected per kilogram of feed or 125 per 100 grams, assuming the feed is completely mixed, the tracer is stable and it meets its specified count.

**Sampling:**

One should take a minimum of four and preferably ten or more "grab" samples from the target batch formulated with the Microtracer. Samples of the target batch should weigh at least 200 grams and ideally be taken at several points in the feed production system. If one analyzes 80 gram "grab" sub-samples of each target sample, one will expect to find 100 tracer particles per analysis. One should then take samples from one or more following batches not formulated with the medication or the tracer. These samples should weigh at least 1-kilo each.

If one analyzes 800 grams of a feed supposed to contain no tracer and finds 100 tracer particles, then if total tracer recovery is 100% of theory one may estimate 10% of the total tracer (and thereby the drug) was found as contamination in that sample. By analyzing very large samples of feeds supposed to contain no tracer, the "sensitivity" (ability of the tracer to allow accurate detection of very low level contamination) may be improved by a factor of 10 or more. Tracer contamination at 0.1% can usually be detected by analyzing 800 gram samples.

Individual batches of feed must be isolated and their flow controlled. People must be ready at each sampling location to take samples as the individual batches pass. All samples must be carefully identified and detailed records of all aspects of the test should be made.

The basic flow of feed at a feedmill is usually as follows:

1. Feed ingredients after being ground to mash with a hammermill (if necessary) are gravity fed into a mixer and mixed.
2. The mixed feed is then dumped into a surge bin.
3. The feed is then discharged from the surge bin using a screw conveyer.
4. The feed is then elevated in the feedmill using a bucket elevator system.
5. The feed is then gravity dumped into a holding bin over a pellet mill.
6. The feed is then pelleted and cooled.
7. The feed is then elevated again using a bucket elevator or pneumatic system.
8. The feed is then distributed to holding bins.
9. The feed is then loaded into trucks.
10. The feed is then carried to farms where it is discharged into bins.
11. The feed is then distributed from the on-farm bins to the animals, poultry or fish for consumption.

Medications are usually added as a microingredient into the mixer, sometimes by hand but often utilizing computer controlled micro-bin systems. "Cross Contamination" of the medication may occur in the micro-bin system even before the medication reaches the mixer. "Cross contamination" to prior batches may also occur, as when mixer discharge gates leak while an earlier batch remains in the surge bin.

As an initial investigation, one may take samples of the "non-target" following feed only at truck loadout taking care to be certain the feed takes the identical route as the medicated feed formulated with Microtracer. If one finds very little or no tracer from the loadout samples, this indicates there may be very little "cross contamination" occurring anywhere in the system. If one finds Microtracers in the "non-target" loadout feed, however, much more extensive testing may be required to diagnose and correct the problem. Portions of the medication may be left as "contamination" to non-target batches at any of the above referenced production points. To reliably diagnose the potential for "cross contamination" of medications to non-target batches of feed, it is critical to take multiple "grab" samples at multiple locations of both the target batch (with medication) as well as one or more following batches. It must be re-emphasized that it is critical the following batches take the identical route through the feedmill as the initial medicated batch formulated with the Microtracer.

Analysis of Samples: Please refer to Microtracer literature items "A-3".

## Results from Actual Tests:

### Test # 1: Premix Plant Manufacturing Powdered Sulfamethazine Premix

Microtracer F-Red was used to estimate contamination of powdered sulfamethazine to flushing materials and to subsequent premix batches. The premix plant had been "red flagged" by the US-FDA because it found more than 5 ppm sulfamethazine in premix supposed to contain none. The premix plant had instituted rigorous flushing cleanout procedures and wanted to confirm these were efficacious.

**Premix Formulation:** 1 % Sulfamethazine (10,000 ppm) and 0.8% Microtracer F-Red (8,000 ppm). Batch size: 2,500-lbs.

| **Batch #1** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 0.41 | 66 | - |
| 2 | 0.40 | 73 | - |
| 3 | 0.47 | 64 | - |
| 4 | 0.38 | 56 | - |
| Average: | 0.42 | 64.75 | 8,800 |
| **% of Specification:** | | **77%** | **88%** |

(Microtracer recovery: 25,000 x 1.66g x 0.8% = 332 formulated count; 259/332 = 77%)

| **Flush #1 – 300lbs of Ground Limestone** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 20.0 | 85 | - |
| 2 | 20.0 | 86 | - |
| 3 | 20.0 | 104 | - |
| 4 | 20.0 | 88 | - |
| Average: | 20.0 | 90.75 | 331 |
| **% of Specification:** | | **2.3%** | **3.3%** |

(Microtracer recovery: 25,000 x 80 x 0.8% = 16,000 formulated count; 363/16,000= 2.3%)

| **Flush #2 – 300lbs of Ground Limestone** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 200.0 | 106 | - |
| 2 | 200.0 | 90 | - |
| 3 | 200.0 | 109 | - |
| 4 | 200.0 | 101 | - |
| Average: | 200.0 | 101.5 | 82 |
| **% of Specification:** | | **0.25%** | **0.82%** |

(Microtracer recovery: 25,000 x 800 x 0.8% = 160,000 formulated count; 406/16,000= 2.3%)

| **Flush #3 – 300lbs of Ground Limestone** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 200.0 | 16 | - |

| | | | |
|---|---|---|---|
| 2 | 200.0 | 30 | - |
| 3 | 200.0 | 14 | - |
| 4 | 200.0 | 27 | - |
| Average: | 200.0 | 21.75 | 65 |
| **% of Specification:** | | **0.054%** | **0.65%** |

| **Flush #4 – 1000lbs of Ground Corn** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 200.0 | 28 | - |
| 2 | 200.0 | 22 | - |
| 3 | 200.0 | 22 | - |
| 4 | 200.0 | 23 | - |
| Average: | 200.0 | 23.75 | 32 |
| **% of Specification:** | | **0.059%** | **0.32%** |

| **Flush #5 – 1000lbs of Ground Corn** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 200.0 | 2 | - |
| 2 | 200.0 | 2 | - |
| 3 | 200.0 | 5 | - |
| 4 | 200.0 | 11 | - |
| Average: | 200.0 | 5 | 10.6 |
| **% of Specification:** | | **0.0125%** | **0.106%** |

| **Follow-Up Batch #1 – 2,500lbs** | | | |
|---|---|---|---|
| Sample # | Sample Size (g) | Microtracer Count | Chemical Assay (ppm) |
| 1 | 200.0 | 4 | - |
| 2 | 200.0 | 2 | - |
| 3 | 200.0 | 1 | - |
| 4 | 200.0 | 2 | - |
| Average: | 200.0 | 2.25 | 1.7 |
| **% of Specification:** | | **0.0056%** | **0.017%** |

**Conclusion:** The various "flushes" did serve a useful purpose in reducing the amount of tracer (and sulfamethazine) reaching premixes supposed to contain none. Contamination was not uniform but rather erratic when present at very low levels. It appeared possible to keep sulfamethazine contamination of following premixes below the objective of 5 ppm.

"Cross Contamination" of the Microtracer F-Red from the premix formulated with the tracer to subsequent flushes and premix batches was less than for the powdered sulfamethazine. It should be possible to make a curvilinear regression to accurately estimate contamination of the drug based upon tracer results. The results evidence the advantage of granulated sulfamethazine over the powdered product as relates to their potential for cross contamination.

Total recovery of the Microtracer F-Red was about 80% of specification and total recovery of the sulfamethazine was about 90%. Most probably, the Microtracer was lower than its specified count. The test continued through four more production batches of premix.

Microtracer was found in all batches and sulfamethazine assays of the final batches were 2.4ppm and 2.5ppm (0.024% and 0.025% of the specified level).

**Test #2: Using Microtracers F to Design a Feedmill, Identifying Locations Where the Greatest Contamination Occurs**

A major designer/supplier of "turnkey" feedmills wanted to minimize batch to batch contamination in their new mills. Microtracer F-Blue was formulated in "Batch #1" at 50 grams per 2,000-lbs of feed into one 3 ton batch (150 grams of tracer mixed in ground corn added to batch). Samples were then taken at the Surge Bin, Conveyer and Top of the Bucket Elevator for five following batches.

**Microtracer F-Blue Contamination to Subsequent Batches:**

| **Batch #2** | | | |
|---|---|---|---|
| Sample # | Surge Bin | Conveyor | Elevator |
| 1 | 2 | 1 | 17 |
| 2 | 1 | 0 | 98 |
| 3 | 1 | 3 | 112 |
| 4 | 0 | 3 | 58 |
| Total: | 4 | 7 | 285 |
| **% of Specification:** | **0.035%** | **0.065%** | **2.59%** |

| **Batch #3** | | | |
|---|---|---|---|
| Sample # | Surge Bin | Conveyor | Elevator |
| 1 | 1 | 3 | 10 |
| 2 | 2 | 0 | 9 |
| 3 | 0 | 0 | 23 |
| 4 | 0 | 1 | 44 |
| Total: | 3 | 4 | 86 |
| **% of Specification:** | **0.018%** | **0.035%** | **0.78%** |

| **Batch #4** | | | |
|---|---|---|---|
| Sample # | Surge Bin | Conveyor | Elevator |
| 1 | 2 | 0 | 2 |
| 2 | 0 | 0 | 0 |
| 3 | 0 | 0 | 0 |
| 4 | 0 | 0 | 15 |
| Total: | 2 | 0 | 17 |
| **% of Specification:** | **0.018%** | **0%** | **0.155%** |

| **Batch #5** | | | |
|---|---|---|---|
| Sample # | Surge Bin | Conveyor | Elevator |
| 1 | 0 | 0 | 0 |
| 2 | 1 | 0 | 0 |
| 3 | 0 | 0 | 1 |

| | | | |
|---|---|---|---|
| 4 | 0 | 1 | 5 |
| Total: | 1 | 1 | 6 |
| **% of Specification:** | **0.009%** | **0.009%** | **0.055%** |

| **Batch #6** | | | |
|---|---|---|---|
| Sample # | Surge Bin | Conveyor | Elevator |
| 1 | 0 | 0 | 1 |
| 2 | 0 | 0 | 1 |
| 3 | 0 | 0 | 1 |
| 4 | 0 | 2 | 6 |
| Total: | 0 | 2 | 9 |
| **% of Specification:** | **0%** | **0.018%** | **0.082%** |

| **Batch #7** | | | |
|---|---|---|---|
| Sample # | Surge Bin | Conveyor | Elevator |
| 1 | 1 | 1 | 0 |
| 2 | 1 | 0 | 0 |
| 3 | 0 | 0 | 129 |
| 4 | 0 | 0 | 0 |
| Total: | 2 | 1 | 129 |
| **% of Specification:** | **0.018%** | **0.009%** | **1.17%** |

**Conclusion:** Comparatively little contamination of the Microtracer occurred at the Surge Bin and Conveyer locations. The amount of contamination was much greater at the top of the bucket Elevator. One sample in Batch #7 contained a "slug" of tracer illustrating that contamination is not uniform and the best chance to find problems is to take and analyze many large samples. The amount of contamination increases as the feed flows through the mill.

In detailed studies at three poultry feed mills (ref. Item "C-9"), the following results were obtained: Recovery of Microtracer F-Red from samples taken from the mixers - 95.7% of specified count

Recovery of Microtracer from pelleted feed samples at truck loading- Feedmill # 1- formula one 82.8%, formula two- 89.3%; Feedmill #2- 75.4%.

Percentage of tracer appearing in non-target feeds- Feedmill # 1- 3.0%, Feedmill #2- 0.26%.

The medicated premix (salinomycin) at Feedmill # 1 was formulated in 60% of the mills feeds whereas the medication (nicarbazin) at feedmill #2 was formulated in only 10% of its production.

Qualitatively, the Microtracer results matched feed medicated assays well, though quantitatively results did not match as well. This was because of variability inherent in the particle statistics used to interpret Microtracer spot counts and more so problems with the medicated feed assays.

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