

PROPER SAMPLING

ONE SIZE DOESN'T FIT ALL!





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IMPORTANT CONCEPTS

- Sampling is predetermined
 - What is the purpose of taking that sample?
 - Method is set, not random—written procedures.
- The sample taken is representative
 - What is your “lot”?
 - Sufficient number and size
- All in the chain of sample collection and handling must have adequate knowledge and training
- Use the right equipment for the job

AN EXAMPLE TO START OUR BRAINS:



**What if we put in the same amount (weight)
of chocolate chips but they were mini?**



What if, in addition to using smaller chocolate chips, we made smaller cookies?



SOME TERMINOLOGY

- The cookie dough is our **matrix**.
- Each cookie is a **sample** of the batch that we made.
- In this case, the thing we are hoping to detect is chocolate chips. These were our **analyte**.

KEY IDEAS

- The **analyte** we are testing for can determine how we sample.
 - They can behave differently
 - Food coloring homogenizes
 - Discrete particles, like chocolate chips, do not
- The **matrix** can also dictate how we sample
 - Static or moving
 - Ground or whole kernel

There are many analytes of interest in grain and feed industries, some impact quality and others to safety

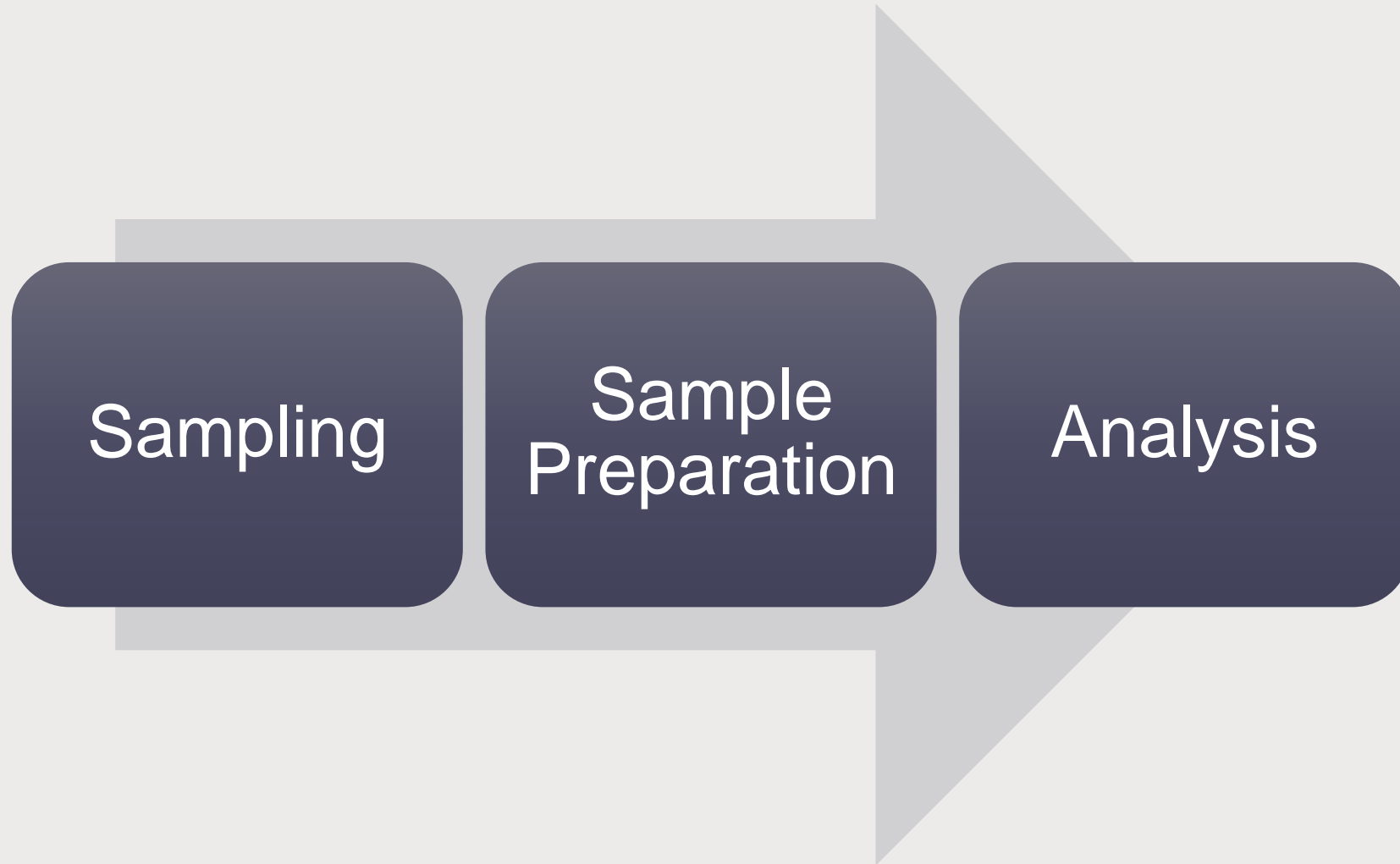
QUALITY EXAMPLES

- Protein
- Oil
- FM
- Moisture
- GM unintended presence

SAFETY EXAMPLES

- Mycotoxins
- *E. coli*
- Pesticide residues
- Veterinary drugs

THREE STEPS IN ANY TESTING PLAN



Sampling plans are designed to overcome as much variability as possible in order to provide a representative sample of the lot for analysis

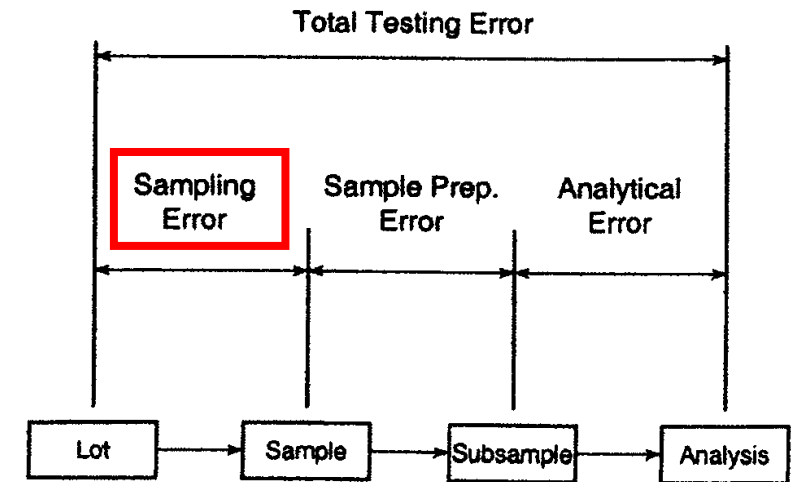


Figure 1. Total variance partitioned into sample, sample preparation, and analytical components.

Johansson, A.S. et. al., C. 2000. Testing Shelled Corn for Aflatoxin, Part I: Estimation of Variance Components. JAOAC Intl. 83:1264-1269.

What are uncertainty and variability?

- Uncertainty: the degree to which a value can (or can't) be considered “true”
- What are the chances that we will grab a sample of cookies and get no chocolate chips?
 - I put 48 chocolate chips into the batch and made 24 cookies, so each will have 2 chocolate chips, right?
- One way uncertainty is expressed is in terms of Variability: the natural variation in some quantity
- How much can the concentration of our analyte (chocolate chips) vary from batch to batch, cookie to cookie, etc...
 - Standard deviation, variance
- Mycotoxins tend to have high variability and therefore, high uncertainty, as would any contaminant that can occur on a single-kernel level

Analytes that are uniformly distributed are less influenced by sampling

- Protein
- Moisture
- Test weight

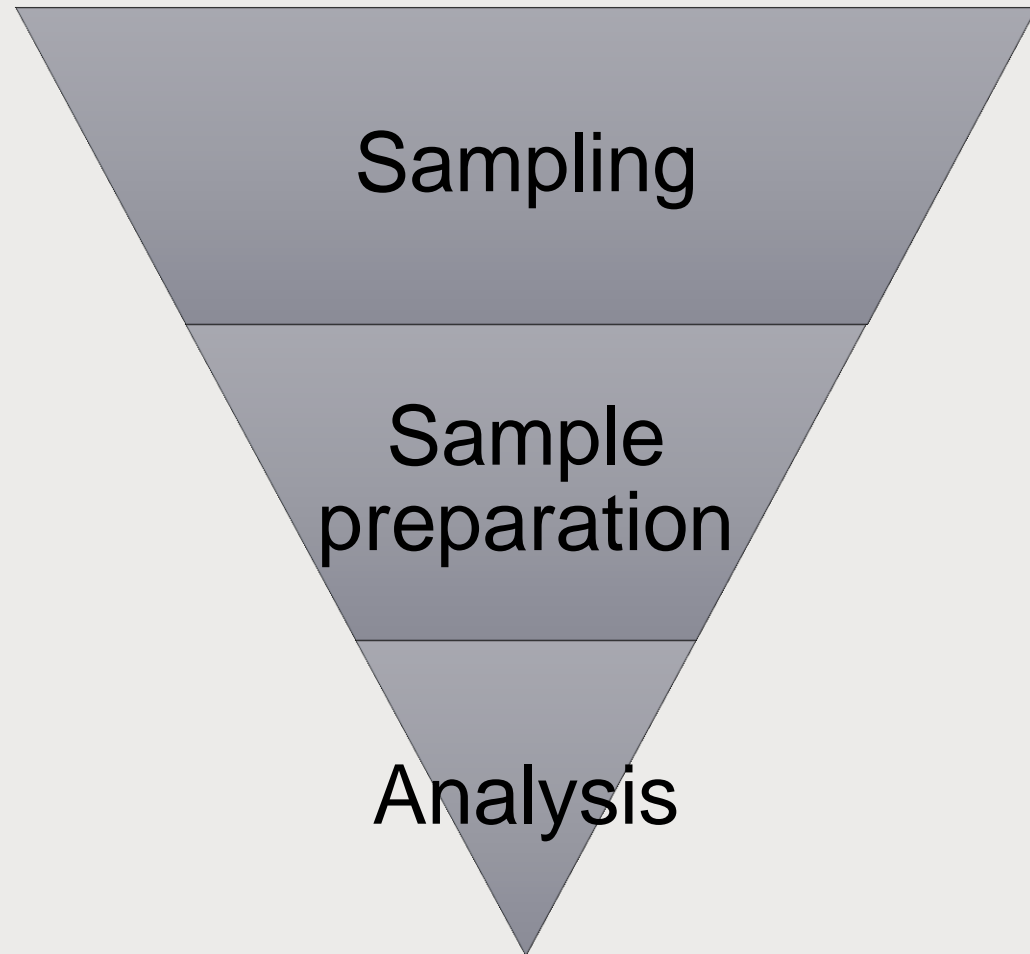
Sampling

Sample
Preparation

Analysis

Analytes that are *not* uniformly distributed tend to be highly influenced by sampling

- Pesticide residues
- Mycotoxins
- Unintended GM
- Foreign material



Non-uniformly distributed analyte example: aflatoxin in corn

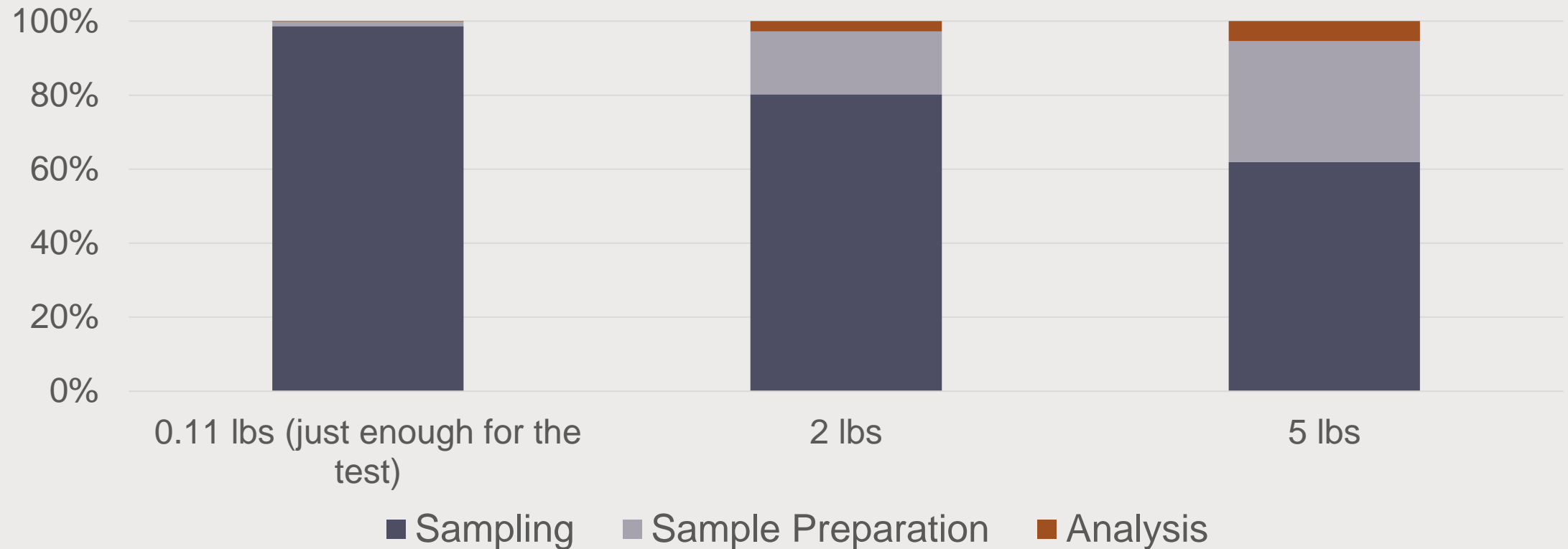
- Documented up to 207,000 ppb aflatoxin in individual corn kernel (62.1 μ g aflatoxin in a 0.3g kernel)
8 of these in a bushel of corn puts it at the 20 ppb limit for use in general commerce
- Johansson et al. study estimated that a shelled corn lot contaminated @ 20 ppb contains appx. 51 aflatoxin contaminated kernels per bushel

Shotwell, O., Goulden, M. and Hesseltine, C. 1974. Aflatoxin: distribution in contaminated corn. Cereal Chem 51:492-499.

Johansson, A.S. et. al., C. 2000. Testing Shelled Corn for Aflatoxin, Part II: Modeling the Observed Distribution of Aflatoxin Test Results. JAOAC Intl. 83:1270-1278

Sampling can significantly effect the variability of a test result

% of aflatoxin test result variability that can be attributed to each step of the testing plan



Obtained using FAO mycotoxin sampling tool. <http://tools.fstools.org/mycotoxins/>

Some analytes are static once they are present in a matrix others can still change

STATIC

- Once the contaminant is in a product it is set; level will not change.
 - Pesticide residues
 - Protein

VARIABLE

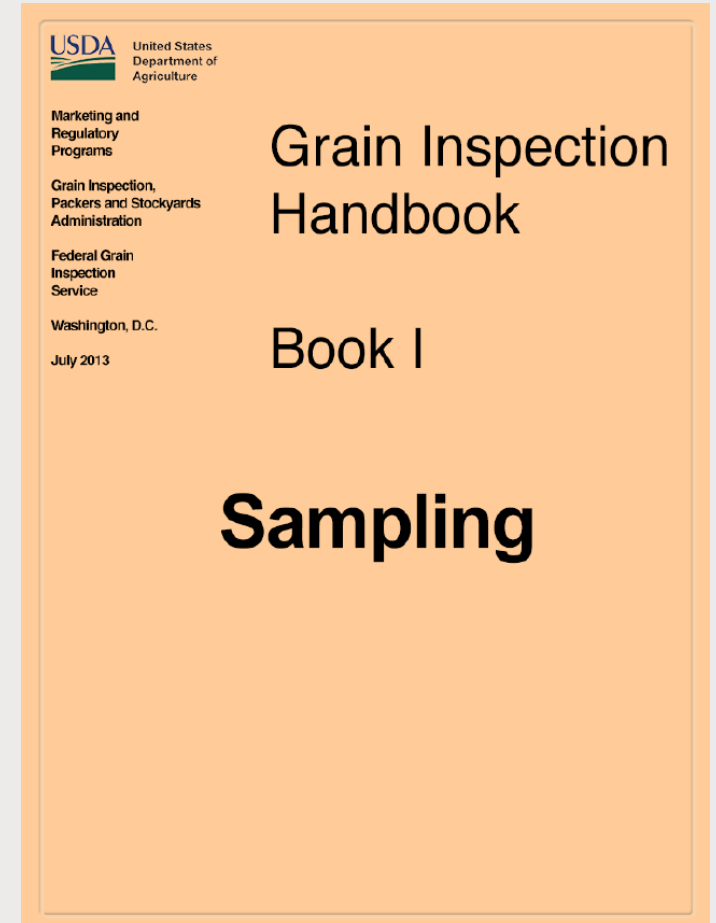
- Once the contaminant is in a product it may change.
 - Microbiologicals
 - Mycotoxins**
 - Sprout
 - Foreign Material

There are prescribed methods for sampling some analytes in some matrices

- FGIS grain inspection handbook outlines strategies for official designations (*e.g., Official Grade*)

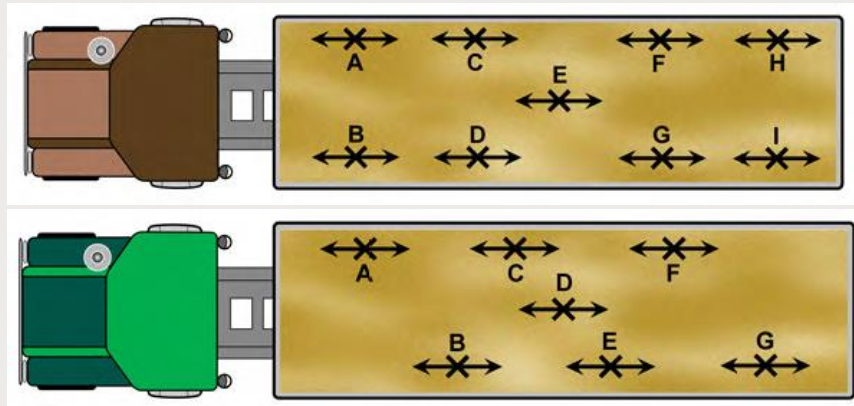
REPRESENTATIVE SAMPLE MUST BE:

- Obtained by official personnel using official procedures and FGIS approved equipment
- Of the prescribed size (appx. 2,500 g)
- Handled securely, protected from manipulation, substitution, and careless handling



From the grain inspection sampling handbook...

- USDA has prescribed sampling methods for various grain transportation units
- Sample representativeness is increased by taking **incremental samples** and aggregating them together
- This is a time-consuming practice
 - May not be practical or feasible to use for every load at some grain facilities
 - Prescribed for official *compliance* sampling (aflatoxin)



Sampling patterns for flat-bottom trucks or trailers containing grain less than 4-feet deep (top) and more than 4-feet deep (bottom)

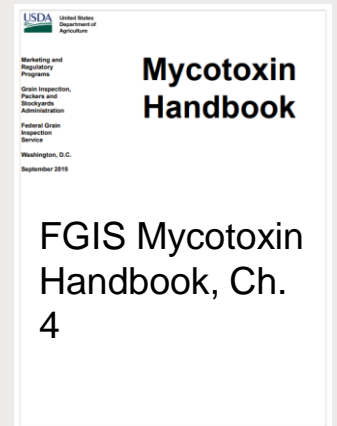
United States Department of Agriculture Federal Grain Inspection Service. 1995. Grain Inspection Handbook - Book 1 Grain Sampling. United States Department of Agriculture Grain Inspection, Packers & Stockyards Administration.

GETTING YOUR SAMPLE

- Incremental samples combined together make the **composite sample**
- The **laboratory sample** is taken from the composite sample
- This sample is then prepared according to needs of analysis that will follow
 - Dividing, grinding, subsampling, etc...
 - The final step of sample preparation will be acquiring the **test portion**

Federal Grain Inspection Service (FGIS) recommended laboratory samples sizes

Commodity	Official Lot Type Minimum sample size (lbs. / grams)			Submitted Samples lbs. / grams
	Trucks or Containers	Railcars	Barges, Sublots & Composite Samples	
Aflatoxin	2 lbs./908 grams	3 lbs./1,362 grams	10 lbs./4,540 grams	10 lbs./4,540 grams (Recommended)
Deoxynivalenol (DON)	200 grams 2 lbs./908 grams (corn only)	200 grams 1000 grams (corn only)	200 grams 1000 grams (corn only)	200 grams 1000 grams (corn only) (Recommended)
Zearalenone	2 lbs./908 grams	3 lbs./1,362 grams	10 lbs./4,540 grams	10 lbs./4,540 grams (Recommended)
Fumonisin	2 lbs./908 grams	3 lbs./1,362grams	10 lbs./4,540 grams	10 lbs./4,540 grams (Recommended)
Ochratoxin A	2 lbs./908 grams	1,000 grams 3 lbs./1,362 grams (corn only)	1,000 grams 10 lbs./4,540 grams (corn only)	1,000 grams 10 lbs./4,540 grams (corn only) (Recommended)



Analysis for grade: Submitted sample minimum=quantity to overflow test weight kettle

Low-level contaminants that are non-uniformly distributed pose a difficult challenge

- Considerations for how to sample for them will likely include:
 - type of business
 - objectives (punishment or knowledge)
 - risk tolerance
 - regulatory compliance
 - market demands
 - other factors

Considerations for development and application of proper sampling practices

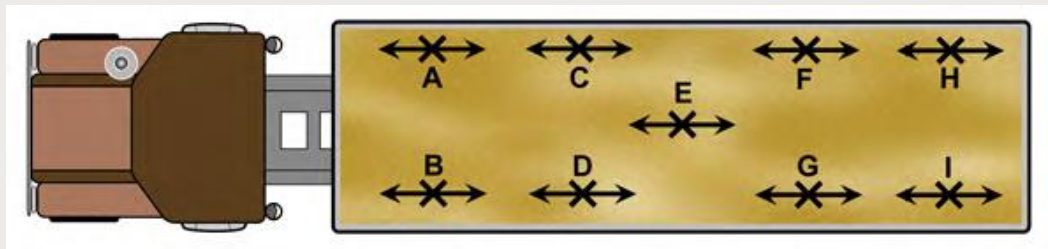


What is the analyte and what is the matrix?

- Analyte: Safety or quality
- Applicable regulations
- Need for traceability, recall capability?
- Market or contract demands/specifications
- Appropriateness of test you will be using
 - FGIS performance-verified mycotoxin test kits (and matrix appropriateness)
 - NTEP approval of moisture meters

Where should the sample be taken?

- Moving or static product
- Ingredient or final product (or both)
- What are you testing for?
- Official or unofficial test



Hurburgh, C.R., Applied Engineering in Agriculture. Vol. 3(1):May, 1987

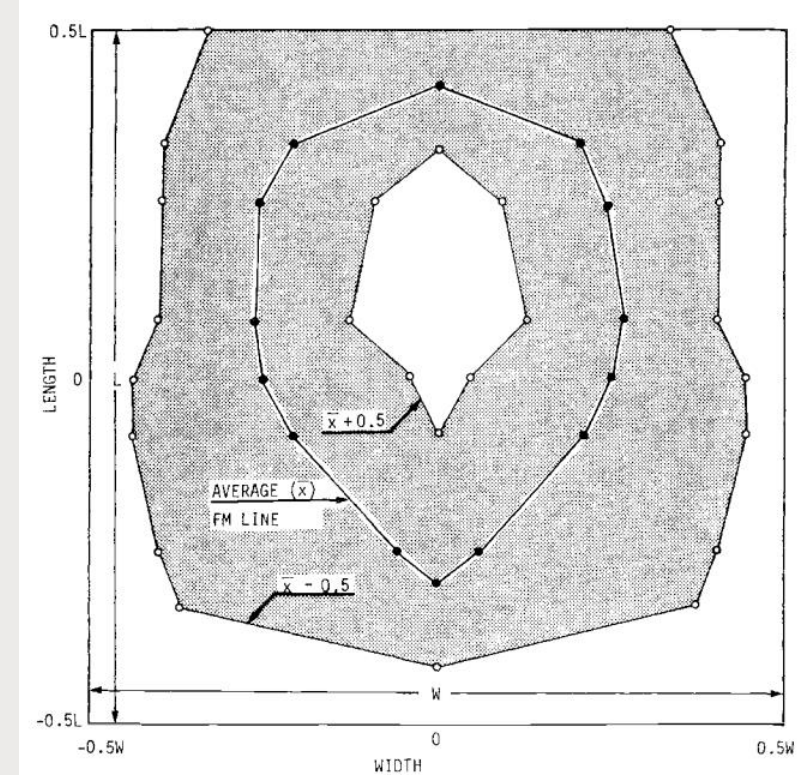


Fig. 2—Truck-sampling locations that contain foreign material within 0.5 percentage point of the load average.

Why is this analysis being performed?

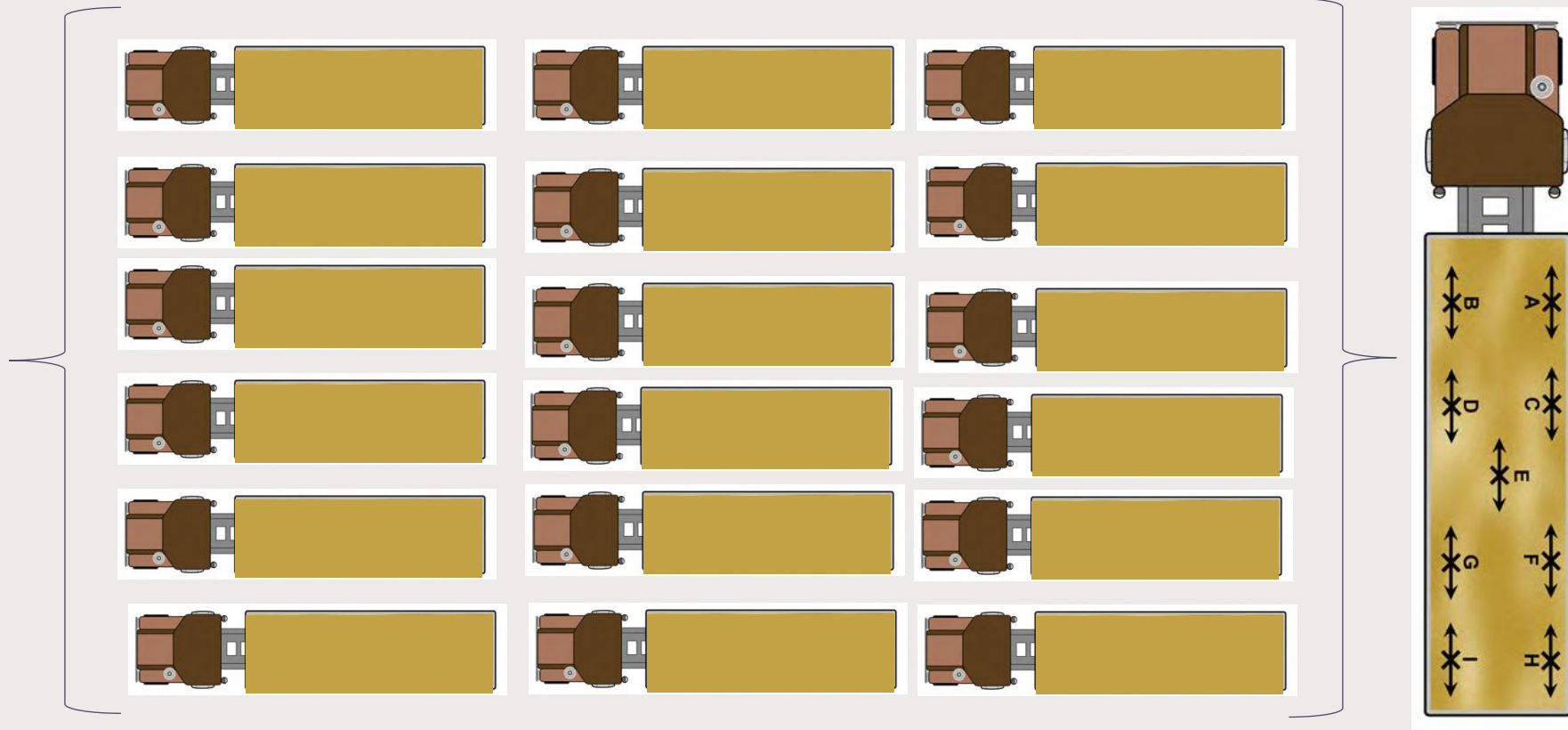
- Market participation
- Reject lots
- Provide COA
- Knowledge of inventory status
- Regulatory compliance (FSMA)
- Documentation of hazard management
- Everyday business practice

When should the sample be taken?

- It depends on what you want to do. Here are three possible ways using the example of hazard management:
 - Control charting - you must sample **inbound ingredients** and **outbound product**
 - Rejection - you must sample at **point of receipt**
 - Inventory knowledge - sample **in-process or upon receipt** (but on aggregate)
- Timeliness of process is important to consider
 - Sampling for uniformly distributed factors poses low risk for any additional “hold up” of workflow than normal, but this is not the case for all analytes.

When should the sample be taken?

- Rejection **point of receipt**
- Inventory knowledge **receipt** (but on aggregate)



How should the sample be taken?

- In a **predetermined, documented manner**
- Consistency in procedures produces usable data
- By a trained individual **(QI)** who understands the implications of *improper* procedure



After sampling comes sample preparation

- Grinding
- Subdivision
- Selection of the test portion



https://www.seedburo.com/pop.asp_Q_potype_E_2_A_imageName_E_3010x.jpg



<https://www.seattlecoffeegear.com/bunn-bulk-commercial-coffee-grinder>



Finally sample analysis

- Different types of tests available, some examples (dependent on matrix and analyte):
 - ELISA, lateral flow, HPLC, PCR, etc...
 - Differences in sensitivity, specificity, cost
 - Each has relevant applicability
- For low level, non-uniformly distributed analytes, these test methods are equally impacted by poor sampling



**The bottom line: a test
result is only as good as the
sample from which it came**

THANK YOU!

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