



The design of appropriate testing plans for testing of genetic purity of a seed lot in the case of a GM variety

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Overview

- What is GM purity testing?
- Comparison between AP testing and GM purity testing
- General assay types for GM purity testing
- SeedCalc Example of GM purity testing
- Questions??



What is GM Purity Testing?

- **Testing Need:** Companies that sell GM seed varieties have a need to deliver seed to the grower with a “high” percentage of the seed with the GM trait
- **Assay is detecting the “success” state rather than the “failure” or deviant state**
- **The definition of “deviant” changes**
 - conforming seed = seed with GM trait
 - deviant seed = conventional seed without the trait
- **Example:** Company claims to deliver seed to grower that has 98% purity for the GM trait (2% is conventional seed or without the trait)



What is GM Purity Testing?

- **Scenario: We have a GM variety lot but we want to test for presence of other unintended GM traits in the GM variety**
- **Is this scenario GM purity testing?**
- **Can we handle this with AP testing approach that Sylvain outlined earlier?**
 - **Deviant(s) become the unintended GM trait(s)**
 - **Assay detects the unintended GM trait**



Comparison between AP testing and GM purity testing



- **Similarities between GM purity and AP testing**
 - Managing tradeoff between consumer and producer risks
 - Definitions of LQL and AQL
 - Use Operating characteristic curves as graphic tool
 - General decision logic is the same
- **How GM purity testing differs from AP testing**
 - Generally constrained to test individual seeds for GM purity testing (measure purity property on each seed)
 - Assay detects the desired state (purity) rather than the undesired state (impurity)
 - Interested in the complement of the AQL and LQL
 - Grower becomes the primary consumer for management of consumer risk (need to meet grower product expectations)



Examples of GM purity testing assays

- **Herbicide Testing**
 - Spray seedlings with herbicide and count the number of surviving seedlings out of the total seedlings
- **Protein Based Testing**
 - ELISA or lateral flow strip systems tested on individual seeds
 - This can be very expensive
- **DNA Based Testing**
 - Run qualitative PCR based method on individual seeds
 - This can be very expensive
 - Other alternates for quantitative PCR?



Use SeedCalc to design testing plan for GM trait purity testing

Example: Company wants to deliver seed lots to grower that have GM trait purity of 98% or better (bag label says 98%)

Seedcalc needs to state things in terms of impurity rather than purity

98% purity standard = 2% impurity



Use SeedCalc to design testing plan for GM trait purity testing



LQL can be defined as 2% impurity (98% GM purity)



Use Seedcalc to find a testing plan to have 95% confidence that accepted seed lots will have purity that exceeds the 98% GM purity standard





Use SeedCalc to design testing plan for GM trait purity testing



LQL entered as 2%

Pool size = 1 for purity testing

SAMPLING TYPE

SINGLE
 DOUBLE

Find Plan
Analyze

Seeds per Pool: 1

false pos. rate	0.0%
false neg. rate	0.0%

1 Stage

Calc 1

# of Pools (N1)	148	% impurity	LQL	AQL
	= 148 seeds		2.00%	0.00%

Opt 1

Accept Lot if # deviants does not exceed: 0 C1

Consumer (beta) Risk	5.03%	Producer (alpha) Risk	0.01%
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Confidence Level (%)

94.97%	99.99%
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Target Consumer Confidence Level: 95.00%

Target Producer Confidence Level: 95.00%

Max # Seeds Tested: 100,000

Test 148 seeds and if all test positive for GM trait then accept the lot

95% confident that accepted lot exceeds 98% GM purity standard

Potential problem: Seed lots at standard of 98% will be rejected 95% of the time!



Use SeedCalc to design testing plan for GM trait purity testing

- Find a better alternative...
- Gather more information
 - Seed production process is capable of producing seed lots with average purity at 99% (Use this as the AQL)
 - Grower does not want seed lots with GM purity below 96% (Use this a LQL)
- Go back to SeedCalc and find another testing plan



Use SeedCalc to design testing plan for GM trait purity testing

IPLING TYPE

SINGLE
DOUBLE

Find Plan
Analyze

Seeds per Pool: 1

false pos. rate	0.0%
false neg. rate	0.0%

1 Stage

of Pools (N1): 260
= 260 seeds

Accept Lot if # deviants does not exceed: 5 C1

LQL	4.00%	AQL	1.00%
Consumer (beta) Risk	5.01%	Producer (alpha) Risk	4.82%
Confidence Level (%)	94.99%		95.18%

Max # Seeds Tested: 100,000

OC Curve Interpretation

Most lots at 4% impurity (96% purity) are rejected

Most lots at 1% impurity (99% purity) are accepted

Test 260 seeds and accept lot if 5 or less seeds are detected without the GM trait



Use SeedCalc to design testing plan for GM trait purity testing

Summary of Testing Plan Results

GM purity level	Probability of accepting lot	Comment
99% (above standard)	95%	High percentage of seed lots will be in this category
98% (at standard)	56%	50/50 chance of accepting lot at standard
96% (below standard)	5%	Sub-standard lots are rejected most of the time





Use SeedCalc to design testing plan for GM trait purity testing



Testing 260 individual seeds with ELISA cost too much...



I can only afford to test 100 seeds.



What can I do? Is it even worth testing?



Look at SeedCalc...





Use SeedCalc to design testing plan for GM trait purity testing

Sampling Type: SINGLE DOUBLE

Find Plan # Seeds per Pool: 1

Analyze false pos. rate: 0.0% false neg. rate: 0.0%

1 Stage

of Pools (N1): 100 = 100 seeds

% impurity: LQL 4.00% AQL 1.00%

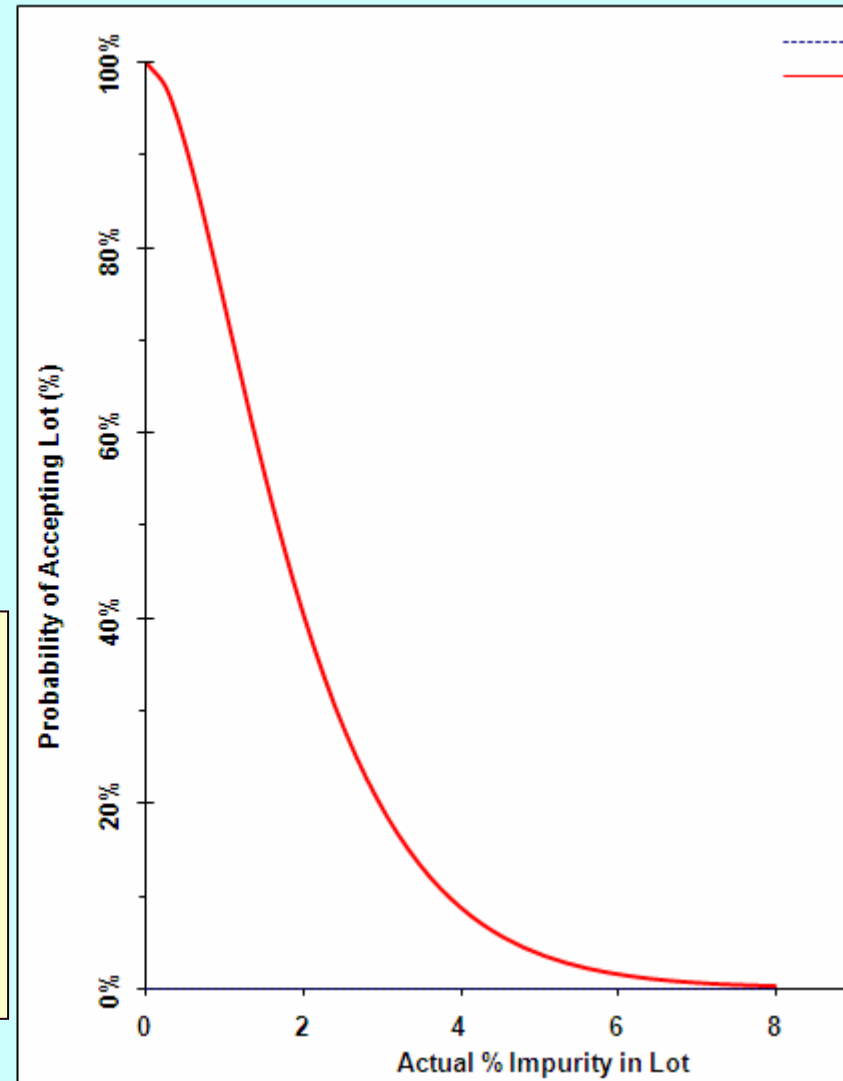
Accept Lot if # deviants does not exceed: 1 C1

Consumer (beta) Risk: 8.72% Producer (alpha) Risk: 26.43%

Confidence Level (%): 91.28% 73.57%

Test 100 seeds and accept lots with one or less seeds without the GM trait

Result: 91% confidence of rejecting sub-standard lot and 74% confident of accepting lot at 99% purity (AQL)





Use SeedCalc to design testing plan for GM trait purity testing

You can also use SeedCalc to estimate purity in a seed lot when you have testing results

Purity Estimation & Confidence Intervals (Assay measures purity characteristic)

(Number of seed sampled should not exceed 10% of total number in population)

# of Individual Seeds Tested	260	% Purity in sample	98.85 %
# Deviants Seeds	3	Desired Confidence Level	95 %
Upper Bound of True % Impurity	2.96	(95% confident that the lot impurity is below 2.96%.)	
2-sided CI for True % Impurity	0.24 to 3.33		
Lower Bound of True % Purity	97.04	(95% confident that the lot purity is above 97.04%.)	
2-sided CI for True % Purity	96.67 to 99.76		





Thank you for your kind attention

Questions?