Using the ICMSF Sampling Plan Tool to assess the performance of a Microbiological Criteria

Part 2: MC for *Listeria monocytogenes* in RTE Foods not supporting growth

Leon G.M. Gorris, PhD

Food Safety Expert, The Netherlands

ICMSF Member 2001-present

Outline

- The Microbiological Criterion (MC) for Ready-to-Eat (RTE) foods that do not support growth of *Listeria monocytogenes* (*Lm*)
- Calculate the performance of the sampling plan with the ICMSF sampling plan tool
- Using the tool for further interpretations of the MC
    - Changing the standard deviation
    - Changing the confidence level

Lm = Listeria monocytogenes MC = Microbiological Criterion RTE = Ready to Eat

MC: RTE foods not supporting Lm growth*

Microbiological criterion for ready-to-eat foods in which growth of *L. monocytogenes* will not occur

Point of application	Microorganism	*n*	*c*	*m*	Class Plan
Ready-to-eat foods from the end of manufacture or port of entry (for imported products), to the point of sale	*Listeria monocytogenes*	5 [a]	0	100 cfu/g [b]	2 [c]

Where *n* = number of samples that must conform to the criterion; *c* = the maximum allowable number of defective sample units in a 2-class plan; *m* = a microbiological limit which, in a 2-class plan, separates acceptable lots from unacceptable lots.

[a] National governments should provide or support the provision of guidance on how samples should be collected and handled, and the degree to which compositing of samples can be employed.

[b] This criterion is based on the use of the ISO 11290-2 method.

Other methods that provide equivalent sensitivity, reproducibility, and reliability can be employed if they have been appropriately validated (e.g., based on ISO 16140).

[c] Assuming a log normal distribution, this sampling plan would provide 95% confidence that a lot of food containing a geometric mean concentration of 93.3 cfu/g and an analytical standard deviation of 0.25 log cfu/g would be detected and rejected based on any of the five samples exceeding 100 cfu/g *L. monocytogenes*.

Such a lot may consist of 55% of the samples being below 100 cfu/g and up to 45% of the samples being above 100 cfu/g, whereas 0.002% of all the samples from this lot could be above 1000 cfu/g.

The typical actions to be taken where there is a failure to meet the above criterion would be to (1) prevent the affected lot from being released for human consumption, (2) recall the product if it has been released for human consumption, and/or (3) determine and correct the root cause of the failure.

*CAC/GL61-2007, "Guidelines on the application of general principles of food hygiene to the control of *Listeria monocytogenes* in foods"

MC: RTE foods not supporting Lm growth

Microbiological criterion for ready-to-eat foods in which growth of *L. monocytogenes* will not occur

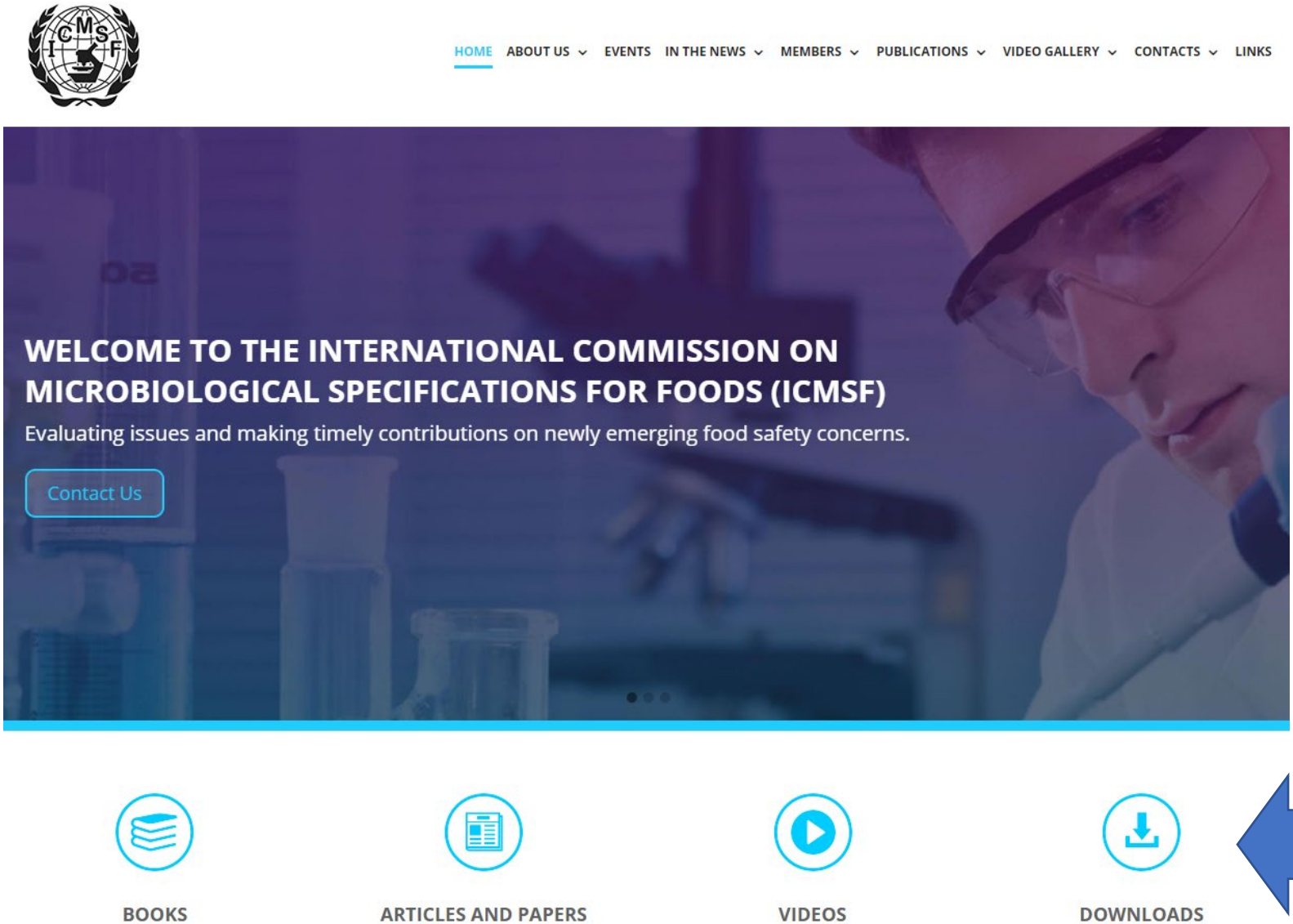
Point of application	Microorganism	*n*	*c*	*m*	Class Plan
Ready-to-eat foods from the end of manufacture or port of entry (for imported products), to the point of sale	*Listeria monocytogenes*	5 [a]	0	100 cfu/g [b]	2 [c]

[c] Assuming a log normal distribution, this sampling plan would provide 95% confidence that a lot of food containing a geometric mean concentration of 93.3 cfu/g and an analytical standard deviation of 0.25 log cfu/g would be detected and rejected based on any of the five samples exceeding 100 cfu/g *L. monocytogenes*.

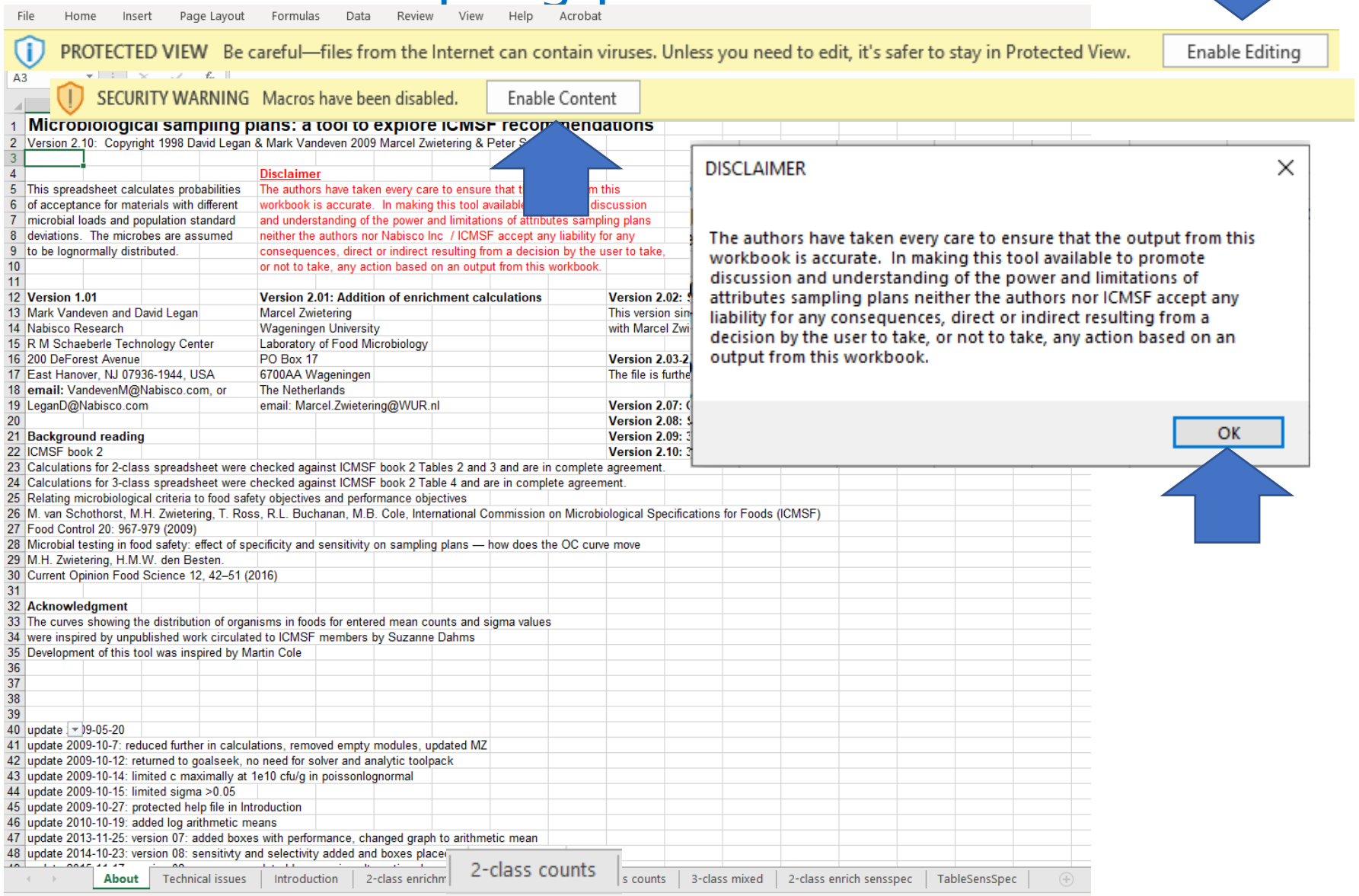
Such a lot may consist of 55% of the samples being below 100 cfu/g and up to 45% of the samples being above 100 cfu/g, whereas 0.002% of all the samples from this lot could be above 1000 cfu/g.

www.ICMSF.org

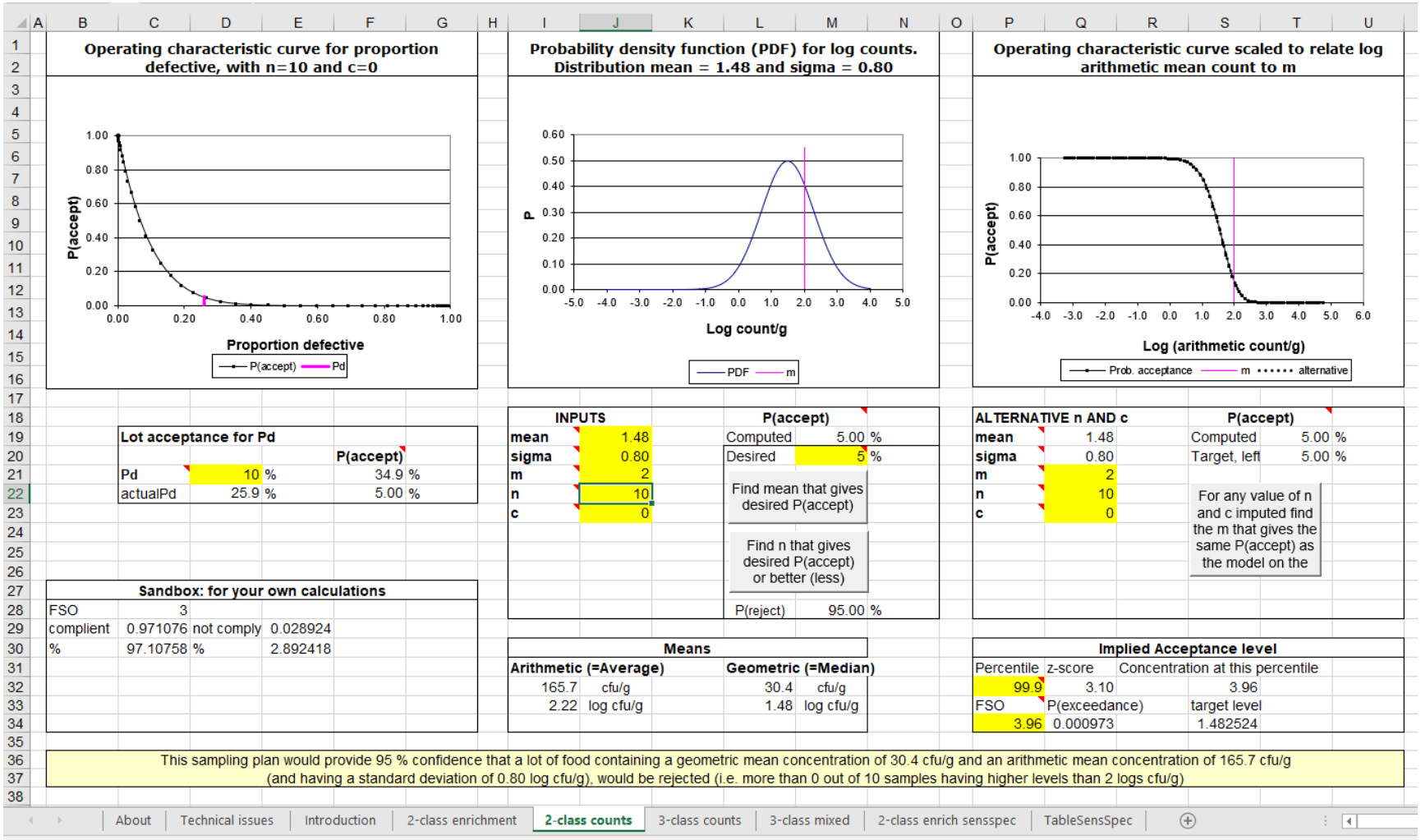
Where to find the tool?



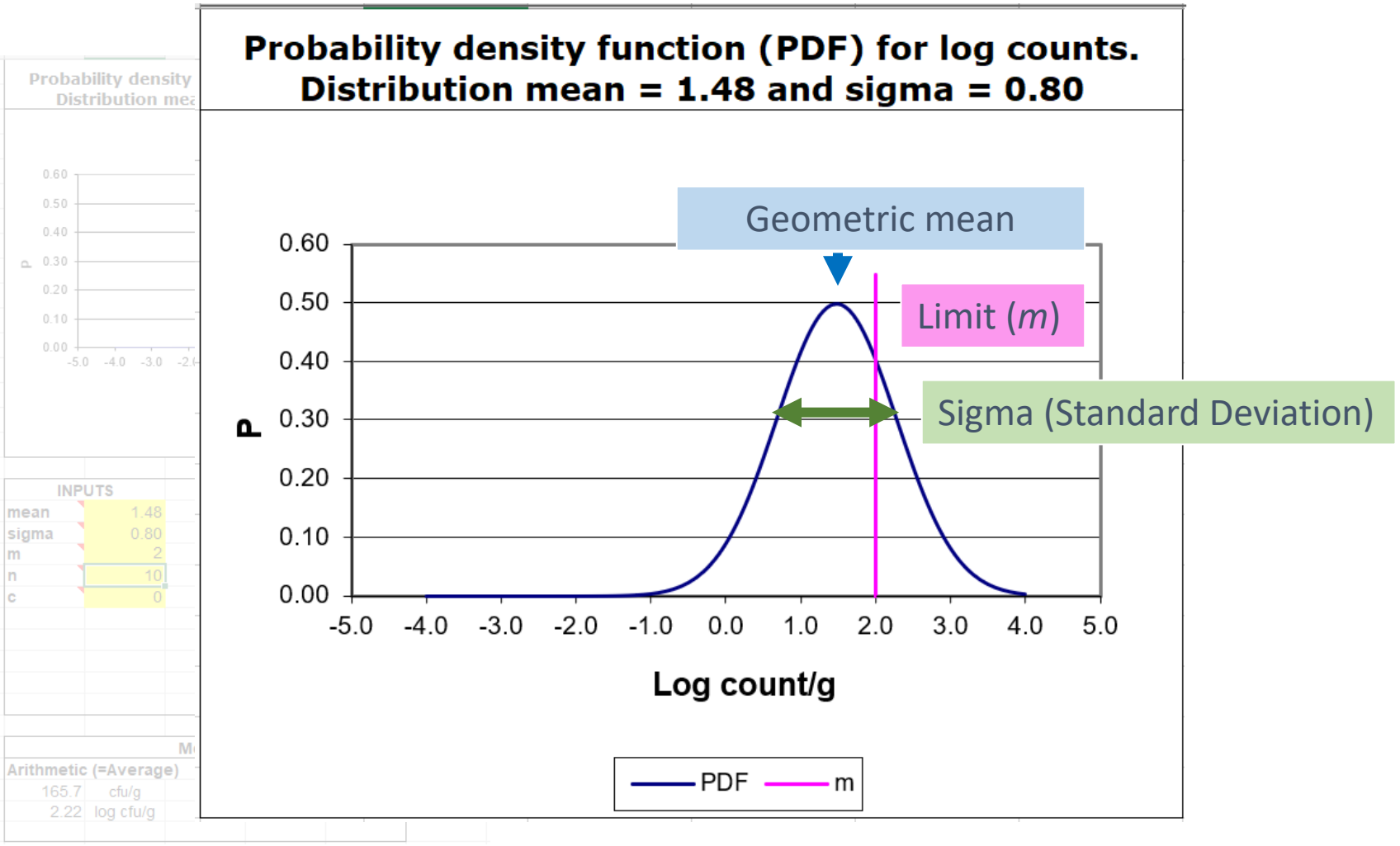
The ICMSF sampling plan tool



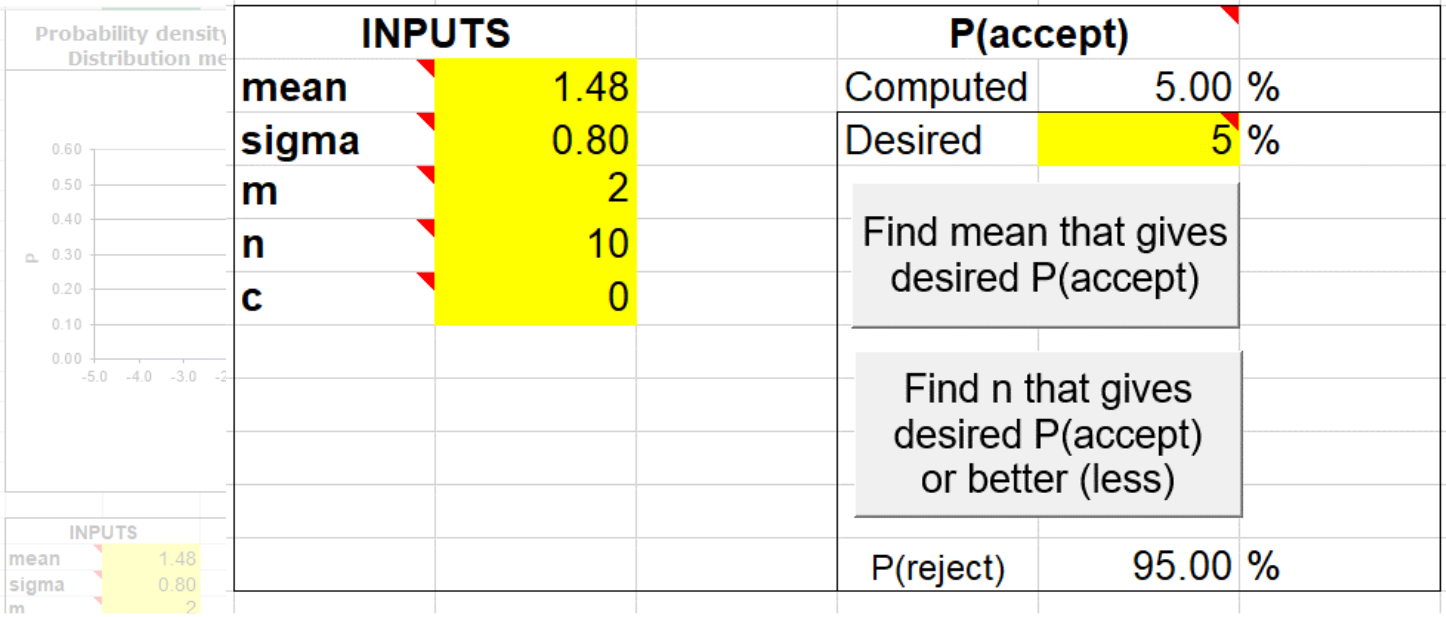
The 2-class counts dashboard



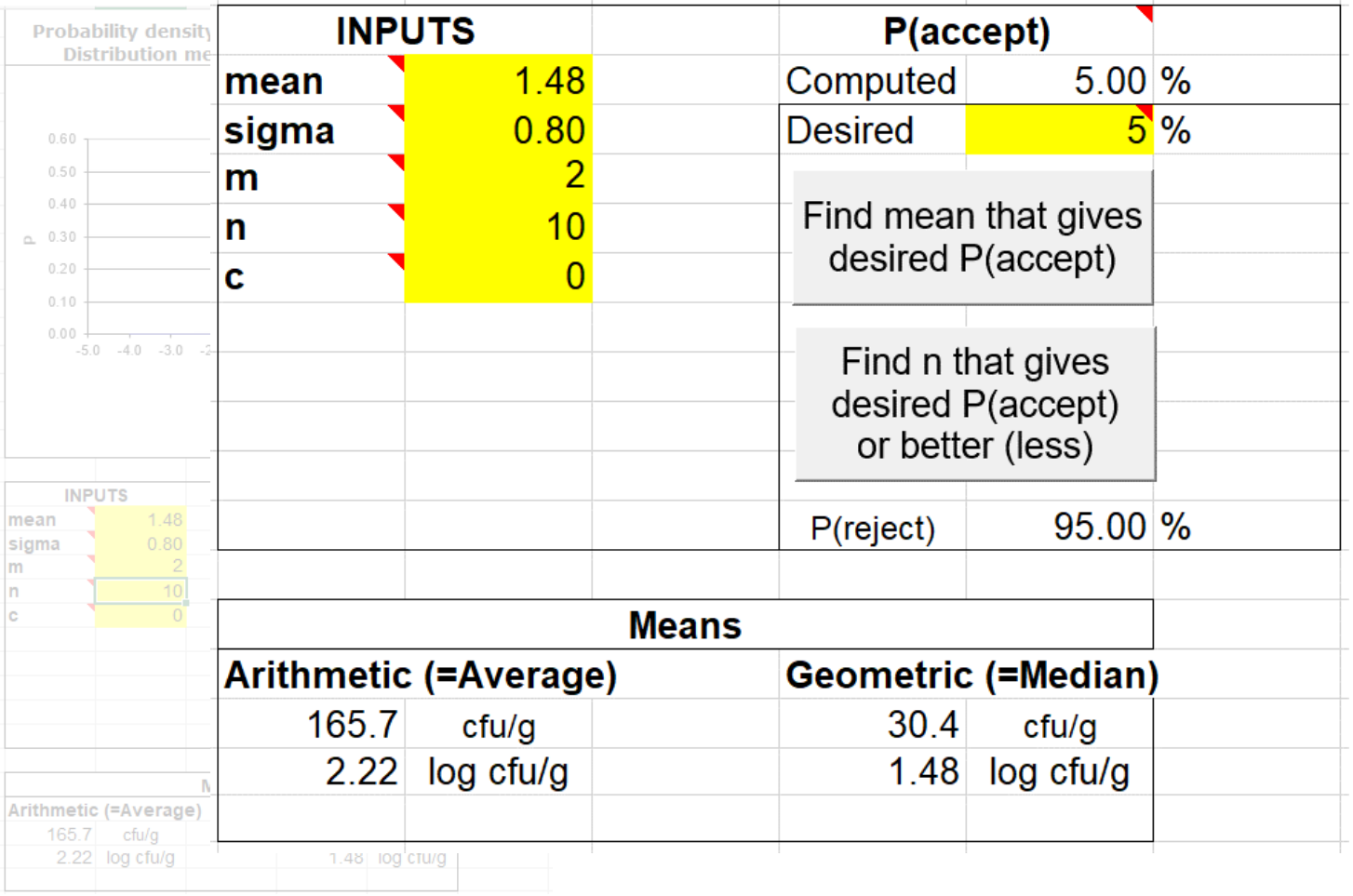
The 2-class counts dashboard



The 2-class counts dashboard



The 2-class counts dashboard



INPUTS	
mean	1.48
sigma	0.80
m	2
n	10
c	0

P(accept)		
Computed	5.00	%
Desired	5	%

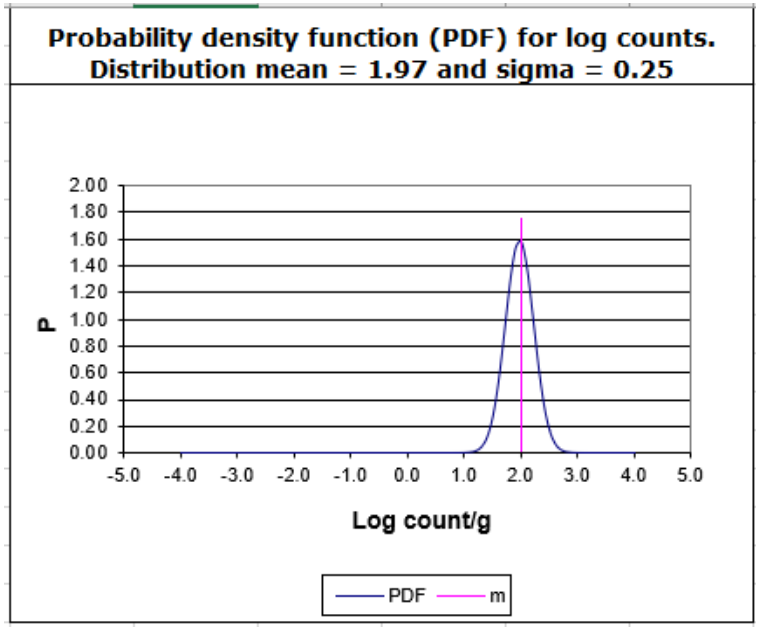
Find mean that gives desired P(accept)

Find n that gives desired P(accept) or better (less)

P(reject)	95.00	%

Means			
Arithmetic (=Average)		Geometric (=Median)	
165.7	cfu/g	30.4	cfu/g
2.22	log cfu/g	1.48	log cfu/g

The Lm MC in the 2-class counts dashboard



Calculated mean and distribution

Standard deviation

n, *c*, *m* from sampling plan

INPUTS		P(accept)	
mean	1.97	Computed	5.00 %
sigma	0.25	Desired	5 %
m	2		
n	5		
c	0		

Find mean that gives desired P(accept)

Find n that gives desired P(accept) or better (less)

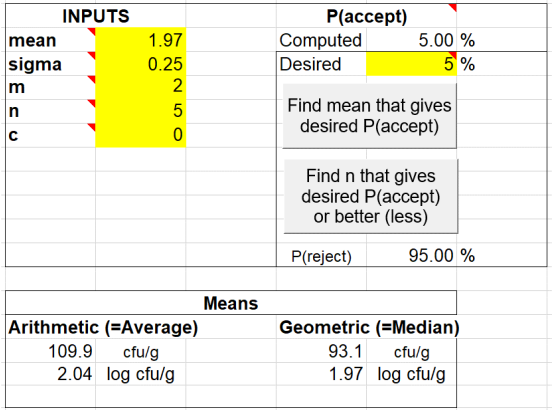
P(reject) 95.00 %

Chance to accept a non-compliant lot

Chance to reject a non-compliant lot

Means			
Arithmetic (=Average)		Geometric (=Median)	
109.9	cfu/g	93.1	cfu/g
2.04	log cfu/g	1.97	log cfu/g

The Lm MC in the 2-class counts dashboard



INPUTS	
mean	1.97
sigma	0.25
m	2
n	5
c	0

P(accept)	
Computed	5.00 %
Desired	5 %

Find mean that gives desired P(accept)

Find n that gives desired P(accept) or better (less)

P(reject) 95.00 %

Means			
Arithmetic (=Average)		Geometric (=Median)	
109.9	cfu/g	93.1	cfu/g
2.04	log cfu/g	1.97	log cfu/g

This sampling plan would provide 95 % confidence that a lot of food containing a geometric mean concentration of 93.1 cfu/g and an arithmetic mean concentration of 109.9 cfu/g (and having a standard deviation of 0.25 log cfu/g), would be rejected (i.e. more than 0 out of 5 samples having higher levels than 2 logs cfu/g)

This sampling plan would provide

- 95% confidence that a lot of food containing a
- geometric mean concentration of 93.1 cfu/g and an
- arithmetic mean concentration of 109.9 cfu/g
- (and having a standard deviation of 0.25 log cfu/g),
- would be rejected
- (i.e. more that 0 out of 5 samples having higher levels than 2 logs cfu/g)

The Lm MC in the 2-class counts dashboard

INPUTS	
mean	1.97
sigma	0.25
m	2
n	5
c	0

P(accept)	
Computed	5.00 %
Desired	5 %

Find mean that gives desired P(accept)

Find n that gives desired P(accept) or better (less)

P(reject) 95.00 %

Means			
Arithmetic (=Average)		Geometric (=Median)	
109.9	cfu/g	93.1	cfu/g
2.04	log cfu/g	1.97	log cfu/g

This sampling plan would provide 95 % confidence that a lot of food containing a geometric mean concentration of 93.1 cfu/g and an arithmetic mean concentration of 109.9 cfu/g (and having a standard deviation of 0.25 log cfu/g), would be rejected (i.e. more than 0 out of 5 samples having higher levels than 2 logs cfu/g)

This sampling plan would provide

- 95% confidence that a lot of food containing a
- geometric mean concentration of 93.1 cfu/g and an
- arithmetic mean concentration of 109.9 cfu/g
- (and having a standard deviation of 0.25 log cfu/g),
- would be rejected
- (i.e. more that 0 out of 5 samples having higher levels than 2 logs cfu/g)

MC: RTE foods not supporting Lm growth

Microbiological criterion for ready-to-eat foods in which growth of *L. monocytogenes* will not occur

Point of application	Microorganism	*n*	*c*	*m*	Class Plan
Ready-to-eat foods from the end of manufacture or port of entry (for imported products), to the point of sale	*Listeria monocytogenes*	5 [a]	0	100 cfu/g [b]	2 [c]

[c] Assuming a log normal distribution, this sampling plan would provide 95% confidence that a lot of food containing a geometric mean concentration of 93.3 cfu/g and an analytical standard deviation of 0.25 log cfu/g would be detected and rejected based on any of the five samples exceeding 100 cfu/g *L. monocytogenes*.

Such a lot may consist of 55% of the samples being below 100 cfu/g and up to 45% of the samples being above 100 cfu/g, whereas 0.002% of all the samples from this lot could be above 1000 cfu/g.

MC: RTE foods not supporting Lm growth

INPUTS	
mean	1.97
sigma	0.25
m	2
n	5
c	0

P(accept)		
Computed	5.00	%
Desired	5	%

Find mean that gives desired P(accept)

Find n that gives desired P(accept) or better (less)

P(reject)	95.00	%

ALTERNATIVE n AND c	
mean	1.97
sigma	0.25
m	2
n	5
c	0

P(accept)		
Computed	5.00	%
Target, left	5.00	%

For any value of n and c imputed find the m that gives the same P(accept) as the model on the

Means			
Arithmetic (=Average)		Geometric (=Median)	
109.9	cfu/g	93.1	cfu/g
2.04	log cfu/g	1.97	log cfu/g

Implied Acceptance level		
Percentile	z-score	Concentration at this percentile
99.9	3.10	2.74
FSO	P(exceedance)	target level
2.00	0.450717	1.225399

level for which the probability of exceedance is calculated

FSO	P(exceedance)
2.00	0.450717

Enter concentration in log cfu/g

MC: RTE foods not supporting Lm growth

INPUTS		P(accept)		
mean	1.97	Computed	5.00	%
sigma	0.25	Desired	5	%
m	2			
n	5	Find mean that gives desired P(accept)		
c	0			
		Find n that gives desired P(accept) or better (less)		
		P(reject)	95.00	%

ALTERNATIVE n AND c		P(accept)		
mean	1.97	Computed	5.00	%
sigma	0.25	Target, left	5.00	%
m	2			
n	5	For any value of n and c imputed find the m that gives the same P(accept) as the model on the		
c	0			

Means		
Arithmetic (=Average)		Geometr
109.9	cfu/g	93.
2.04	log cfu/g	1.9

f_x =1-NORM.DIST(P34,meanlog,sigma,1)

Tool calculates P(exceedance)

FSO	P(exceedance)
2.00	0.450717

MC: RTE foods not supporting Lm growth

Concentration (cfu/g)	P(exceed) calculation	Proportion of food lot over chosen concentration
10	FSO 1.00 P(exceedance) 0.999947	99.99 %
100	FSO 2.00 P(exceedance) 0.450717	45.07 %
500	FSO 2.70 P(exceedance) 0.001729	0.17 %
1000	FSO 3.00 P(exceedance) 1.86E-05	0.00186 %
5000	FSO 3.70 P(exceedance) 2.2E-12	0.00000000022 %

MC performance: Changing assumptions

Microbiological criterion for ready-to-eat foods in which growth of *L. monocytogenes* will not occur

n	*c*	*m*	Class Plan
5 [a]	0	100 cfu/g [b]	2 [c]

Standard Deviation (SD) = sigma = 0.25 log cfu/g

Confidence Level (CL) = P(reject) = 95%

Changing assumptions: standard deviation (SD)

Sigma
0.25
0.50
0.80
1.20
1.50

INPUTS		P(accept)		
mean	1.97	Computed	5.00	%
sigma	0.25	Desired	5	%
m	2	Find mean that gives desired P(accept)		
n	5			
c	0	Find n that gives desired P(accept) or better (less)		
		P(reject)	95.00	%

Means			
Arithmetic (=Average)		Geometric (=Median)	
109.9	cfu/g	93.1	cfu/g
2.04	log cfu/g	1.97	log cfu/g

Confidence Level (CL) is 95%

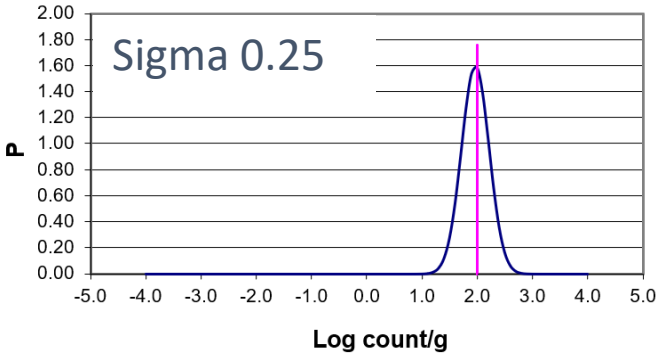
Sigma represents Standard Deviation (SD)

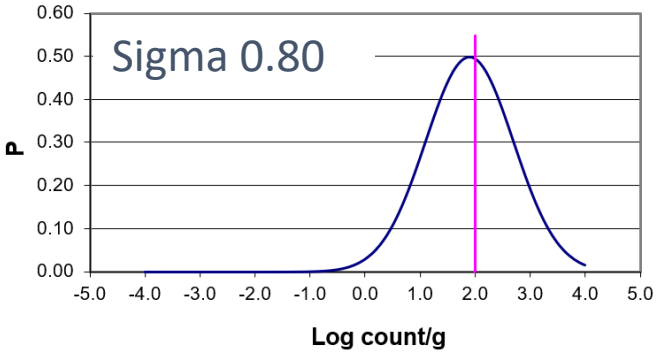
Changing assumptions: standard deviation (SD)

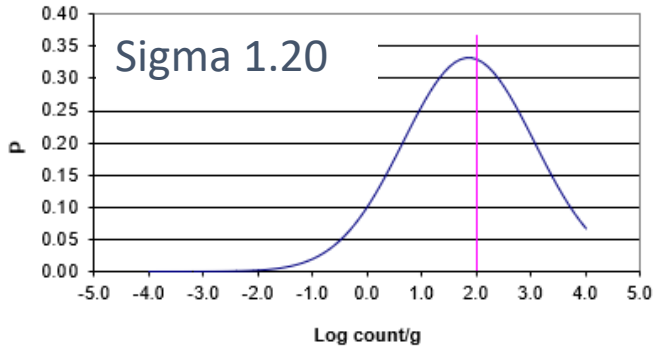
Sigma
0.25
0.50
0.80
1.20
1.50

INPUTS		P(accept)	
mean	1.97	Computed	5.00 %
sigma	0.25	Desired	5 %
m	2	Find mean that gives desired P(accept)	
n	5	Find n that gives desired P(accept) or better (less)	
c	0	P(reject)	95.00 %

Means			
Arithmetic (=Average)		Geometric (=Median)	
109.9	cfu/g	93.1	cfu/g
2.04	log cfu/g	1.97	log cfu/g







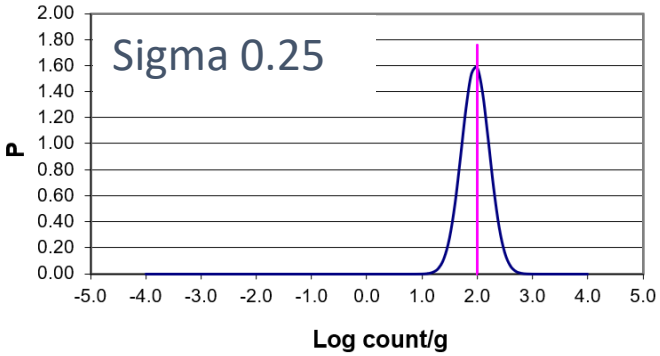
Confidence Level (CL) is 95%

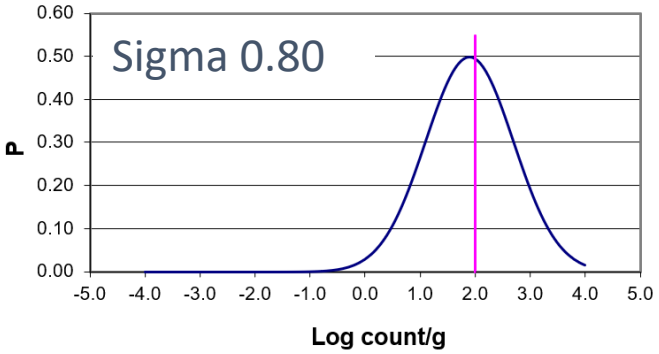
Sigma represents Standard Deviation (SD)

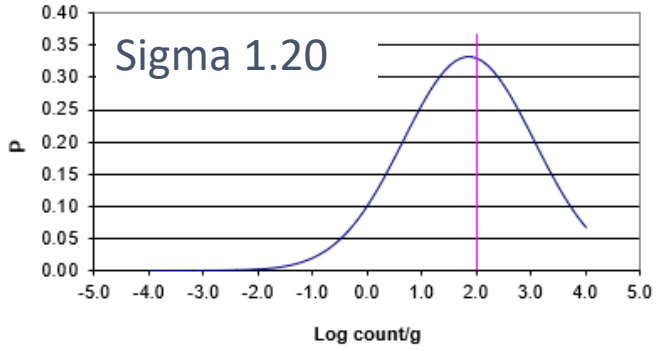
Changing assumptions: standard deviation (SD)

Sigma	Arithmetic mean		Geometric mean	
log cfu/g	*cfu/g*	*log cfu/g*	*cfu/g*	*log cfu/g*
Codex **0.25**			93.1	1.97
0.50			86.7	1.94
0.80			79.6	1.90
1.20			71.0	1.85

Limit *m* = 2 log cfu/g







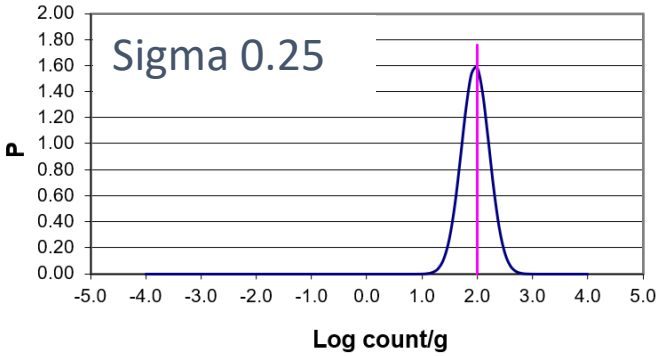
Confidence Level (CL) is 95%

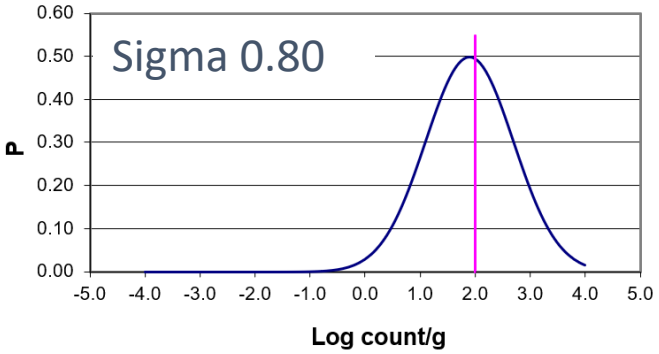
Sigma represents Standard Deviation (SD)

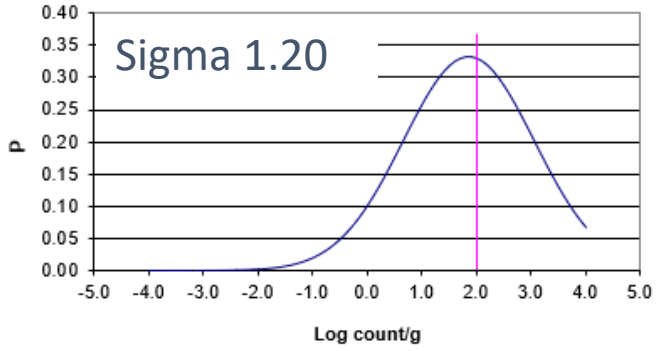
Changing assumptions: standard deviation (SD)

Sigma	Arithmetic mean		Geometric mean	
log cfu/g	*cfu/g*	*log cfu/g*	*cfu/g*	*log cfu/g*
Codex **0.25**	110	2.04	93.1	1.97
0.50	168	2.23	86.7	1.94
0.80	434	2.64	79.6	1.90
1.20	3230	3.51	71.0	1.85

Limit *m* = 2 log cfu/g







Confidence Level (CL) is 95%

Sigma represents Standard Deviation (SD)

Impact of sigma on lot status

Calculate using P(exceedance)

Sigma	Estimated Lm levels in proportions (%) of a food lot				
(log cfu/g)	≤ 100 (2 log)	> 100 (2 log)	> 1000 (3 log)	> 10000 (4log)	> 100000 (5 log)
Codex **0.25**	55 %	45 %	0.002 %	0.00000	0.00000
0.80	55 %	45 %	8.5 %	0.43 %	0.00536 %
1.20	55 %	45 %	16.9 %	3.67 %	0.435 %

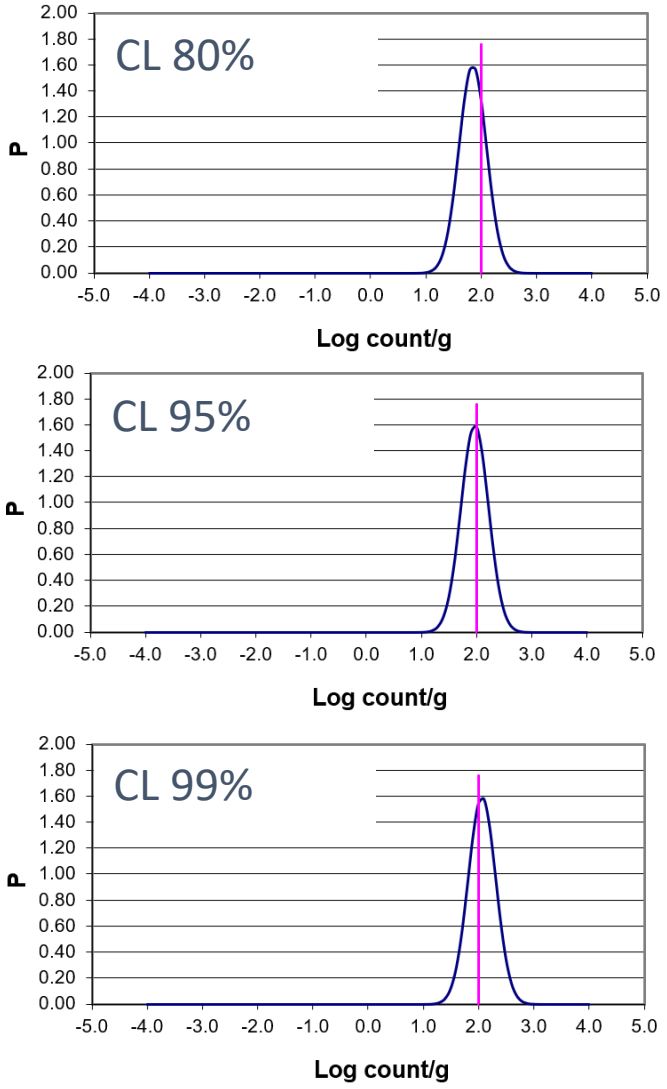
Confidence Level (CL) is 95%

Sigma represents Standard Deviation (SD)

Changing assumptions: confidence level

Confidence	Arithmetic mean		Geometric mean	
Level (%)	*cfu/g*	*log cfu/g*	*cfu/g*	*log cfu/g*
80.00			70.90	1.85
90.00			82.50	1.92
Codex **95.00**			93.10	1.97
99.00			116.00	2.06
99.90			147.00	2.17

Limit *m* = 2 log cfu/g



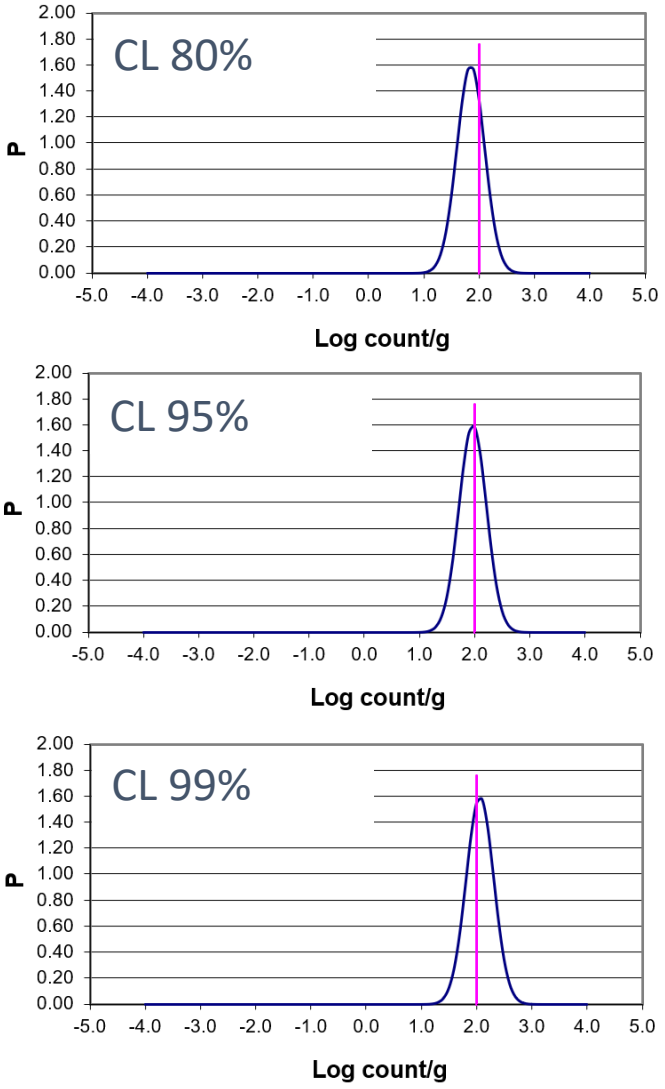
Standard Deviation is 0.25 log cfu/g

Confidence Level (CL) represents P(reject)

Changing assumptions: confidence level

Confidence	Arithmetic mean		Geometric mean	
Level (%)	*cfu/g*	*log cfu/g*	*cfu/g*	*log cfu/g*
80.00	83.70	1.92	70.90	1.85
90.00	97.40	1.99	82.50	1.92
Codex **95.00**	109.90	2.04	93.10	1.97
99.00	136.90	2.14	116.00	2.06
99.90	173.50	2.24	147.00	2.17

Limit *m* = 2 log cfu/g



Standard Deviation is 0.25 log cfu/g

Confidence Level (CL) represents P(reject)

Changing confidence level and standard deviation

Sigma	Confidence	Arithmetic mean		Geometric mean	
log cfu/g	**Level (%)**	cfu/g	log cfu/g	cfu/g	log cfu/g
0.25	**80.00**				
0.25	**90.00**				
0.25	**95.00**	109.90	2.04	93.10	1.97
0.80	**80.00**				
0.80	**90.00**				
0.80	**95.00**				
1.20	**80.00**				
1.20	**90.00**				
1.20	**95.00**				

Codex

Limit *m* = 2 log cfu/g

Standard Deviation is 0.25 log cfu/g

Confidence Level (CL) represents P(reject)

Changing assumptions: confidence level

Sigma	Confidence	Arithmetic mean		Geometric mean	
log cfu/g	**Level (%)**	cfu/g	log cfu/g	cfu/g	log cfu/g
0.25	**80.00**	83.70	1.92	70.90	1.85
0.25	**90.00**	97.40	1.99	82.50	1.92
0.25	**95.00**	109.90	2.04	93.10	1.97
0.80	**80.00**	181.60	2.26	33.30	1.52
0.80	**90.00**	294.70	2.47	54.00	1.73
0.80	**95.00**	434.30	2.64	79.60	1.90
1.20	**80.00**	873.70	2.94	19.20	1.28
1.20	**90.00**	1805.50	3.26	39.70	1.60
1.20	**95.00**	3230.80	3.51	71.00	1.85

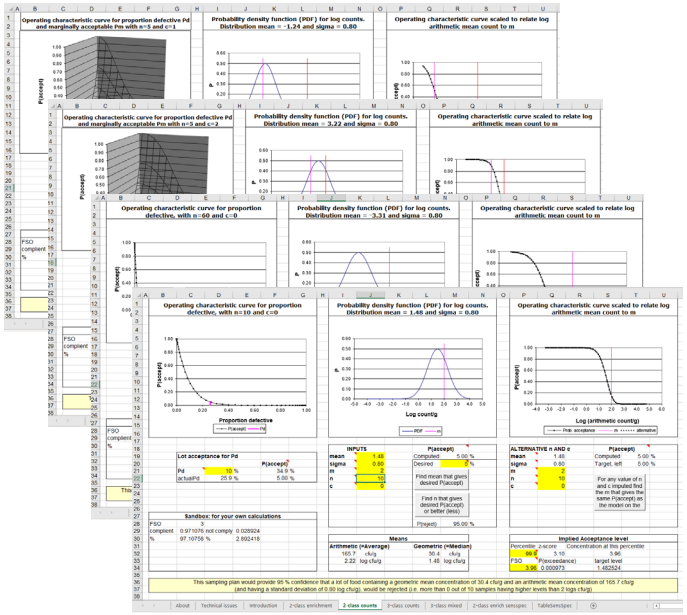
Codex

Limit *m* = 2 log cfu/g

Standard Deviation is 0.25 log cfu/g

Confidence Level (CL) represents P(reject)

Summary



- The ICMSF sampling plan tool can be used to calculate and further interpret the performance of sampling plans included in Microbiological Criteria
- The robustness of key assumptions should be understood and choices should be well informed.
- See our other clips to understand how the ICMSF Sampling plan tool can be used to assess and interpret the performance of these MCs

For more information, see www.icmsf.org