Feed Additives – What research do you need to assess their value?

Bill Stone and Gerald Poppy

Diamond V
The Trusted Experts In Nutrition & Health®

What is Evidence Based Medicine?

- Evidence based medicine (EBM) was originally defined as the conscientious, explicit, and judicious use of current best evidence in making decisions about the care of individual patients.
- The practice of evidence based medicine means integrating individual clinical expertise with the best available external clinical evidence from systematic research.

- (Sackett DL, Rosenberg WMC, Gray JAM, Haynes RB, Richardson WS. Evidence based medicine: what it is and what it isn't. BMJ 1996; 312: 71-2)

Diamond V

Why might it be relevant to you?

- The process for making a decision to “intervene,” whether for treating a sick cow or buying a piece of equipment or implementing a feeding practice, can be approached in a systematic way.

Updated definition

- **Evidence-Based Practice** is defined as, "Making a conscientious effort to base clinical decisions on research that is most likely to be free from bias, and using interventions most likely to improve how long or well patients live."
  (Mark H. Ebell, MD, MS, Professor, University of Georgia, Editor-in-Chief, Essential Evidence Plus)
- What is the process for using “evidence” to make decisions

Diamond V

Two categories of information

- Risk – What is the probability the outcome will be what I expect.
- Finance – What is the expected payout in terms of money at risk and profit, (taking into account the risk)

Diamond V

All Information is not created equal

- How do you rank the “validity” of the research
  - Does it target the right population
  - How well does it control for “confounding and bias”
  - What does the “stats” tell you

Diamond V

Table 15.1 Hierarchy of evidence.

Hierarchy of evidence	General description	Methodology	Description
Strongest	Overview	*Systematic review and meta-analysis*	*Systematic review*: a literature review focused on a single question which tries to identify, appraise, select and synthesize all high quality research evidence relevant to that question. *Meta-analysis*: a systematic review that uses quantitative methods to synthesize and summarize the results (see Section 13.5).
	Experimental study	*Randomized controlled trial*	Animals are randomly allocated into one or more experimental groups or a control group and followed over time for the variables/outcomes of interest (see Sections 5.3–5.9).
	Observational study	*Cohort study*	Groups (cohorts) of animals are selected on the basis of their exposure to a particular factor and followed prospectively to see if they incur a specific outcome (e.g. disease) – see Section 5.2.3(a).
		Case-control study	Animals which have the outcome of interest (cases) and those that do not have the same outcome (controls) are identified, and a retrospective analysis performed to see if there are differences between the groups in their exposure(s) of interest – see Section 5.2.3(b).
		Cross-sectional survey	A defined population is observed at a single point in time or time interval. Exposure and outcome are determined simultaneously.
	Observation	*Case series and case report*	A report based on a single animal; sometimes collected together into a short series but there are no controls.
	Opinion	*Expert opinion*	A view or consensus of views from one or more persons with a high degree of skill in or knowledge of the relevant subject.
Weakest		*Anecdotal*	Based on casual observations or indications rather than rigorous or scientific analysis.

Statistics for Veterinary and Animal Science, 2nd ed., Petrie and Watson, Blackwell Pub (2006) pg. 215

Diamond V

http://library.downstate.edu/EBM2/2100.htm

Diamond V

Evidence Hierarchy

Adapted from Figure 4 – Relative strengths of evidence provided by different methods used in clinical research illustrated diagrammatically in the so-called pyramid of evidence. Strength of association increases from the base to the peak of the pyramid.

JAVMA, Vol 235, No. 9

Case Reports

- Weak evidence because no control of confounders, (change in weather, feed, DIM of cows, parity, etc.)
- Control group not randomized, (another method of controlling confounding and bias.)

Diamond V

What are case reports

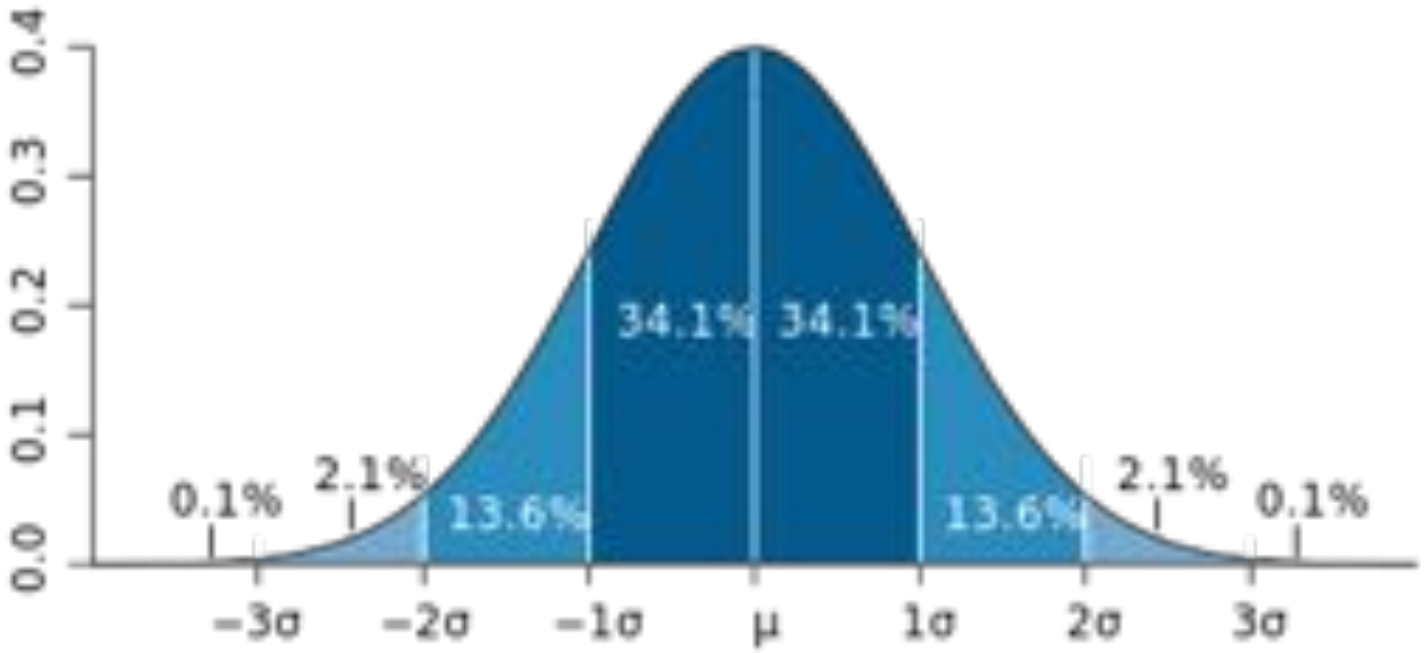
- **What are case reports?**
  - **Case series and Case reports consist of collections of reports on the treatment of individual patients or a report on a single patient. Because they are reports of cases and use no control groups to compare outcomes, they have little statistical validity.**
- **Example**
  - **I fed X to the dairy and thought I saw a decrease in death and GI upsets. I took it out and things went back to where they were.**
  - **Steve started feeding something the IBA guy is selling. The vet says the manure is better, and Steve thinks the cows may be up in milk**
  - **We think feed efficiency improved when we fed Diamond V.**

Diamond V

Other examples of case reports

- Blood letting prior to 1900 was supported by a lot of really smart scientists. (Benjamin Rush)
- They relied on their clinical judgment.
- Not necessarily bad, just not as accurate
- Development of the Koch postulates and later Hills postulates, development of causal theory.

The **standard error of the mean** (SEM)



- **Normal distribution of sample mean relative to the true mean from a sample estimate of the population.**
- **Do we know what the "True Mean" is?**
- **If we resample the population, where will the "mean" of the new sample fall?**

Diamond V

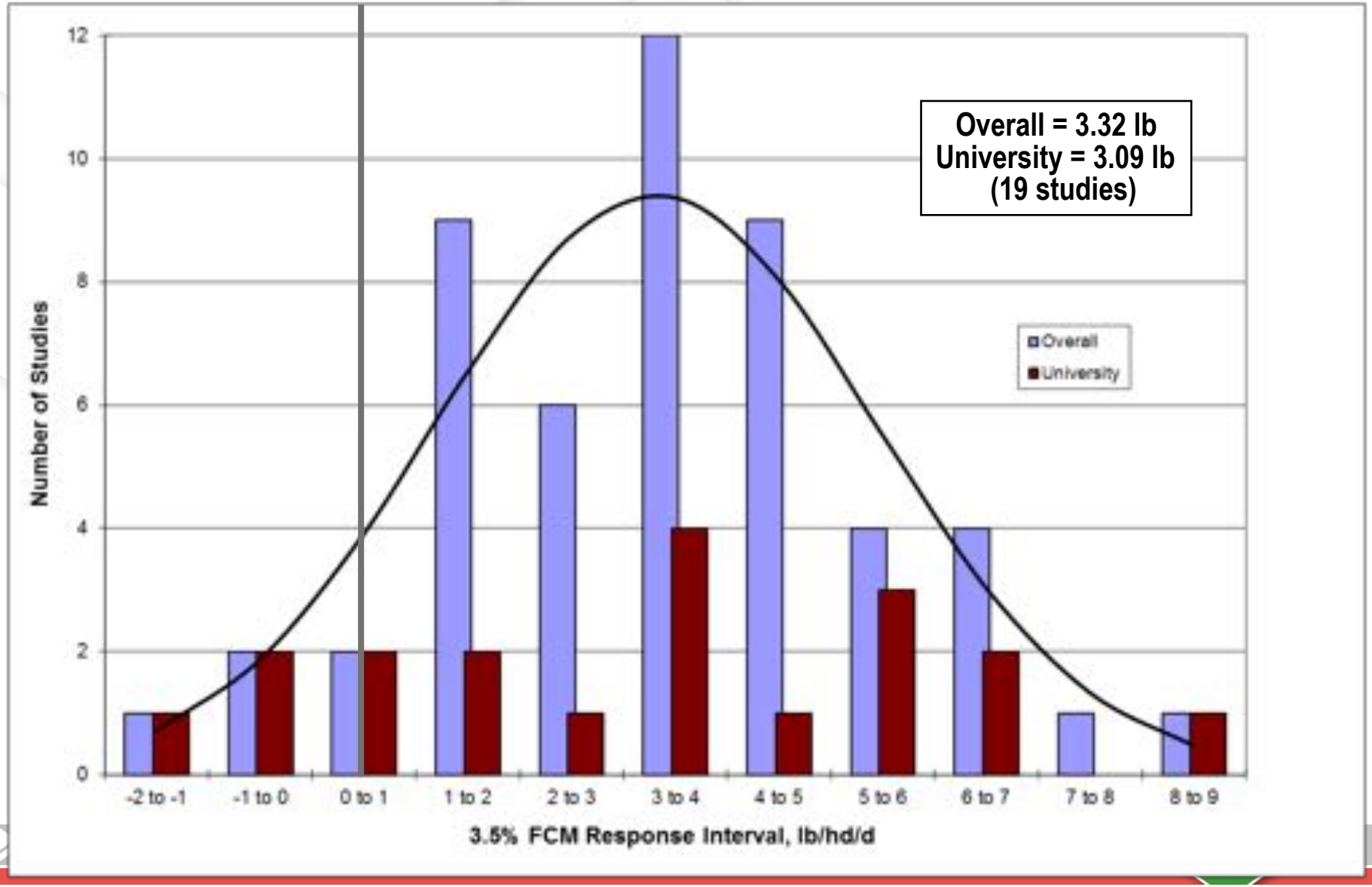
Standard error of the mean

$$\mathrm{SE}_{\bar{x}} = \frac{s}{\sqrt{n}}$$

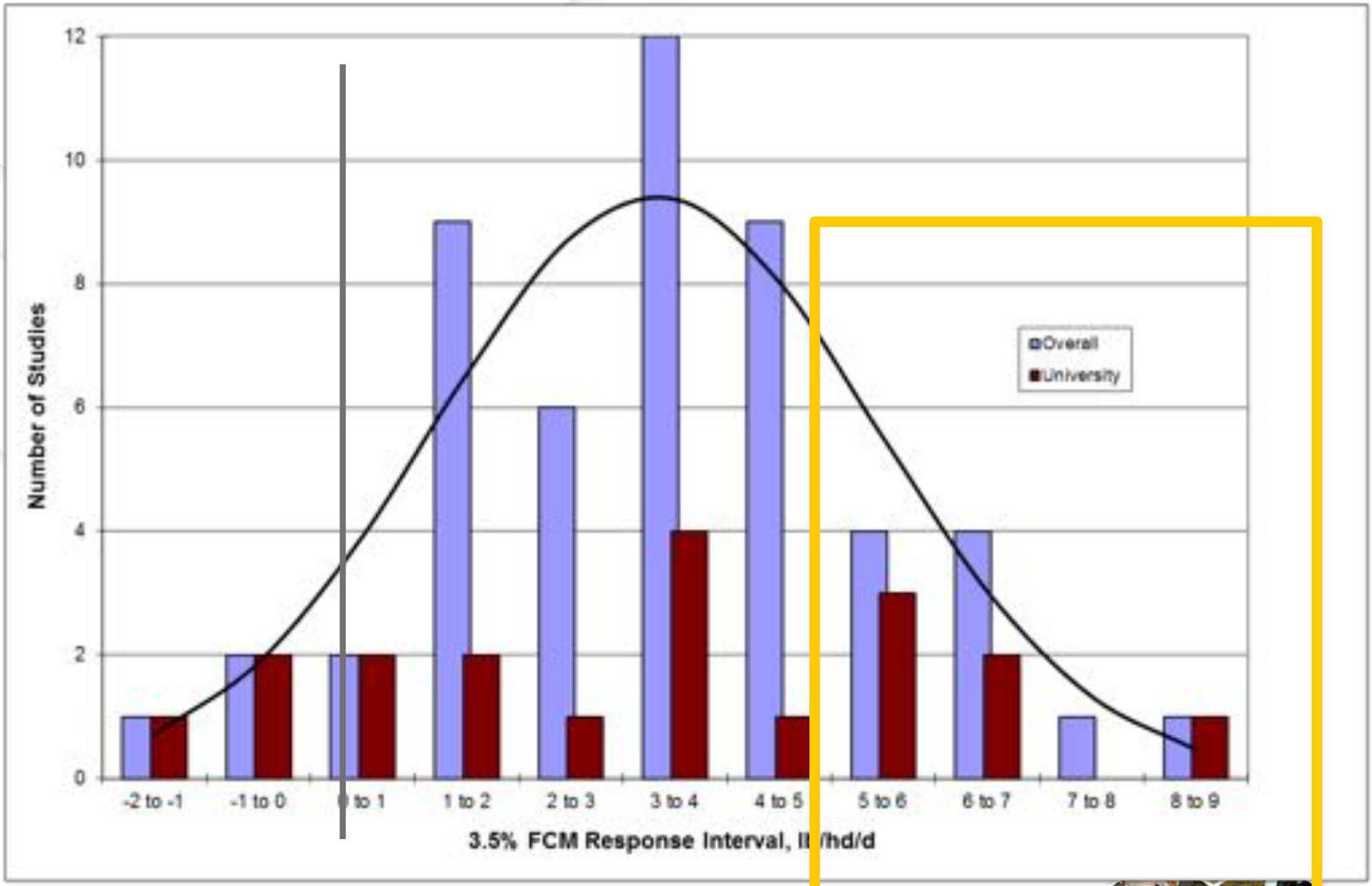
What does the formula tell us?

Diamond V

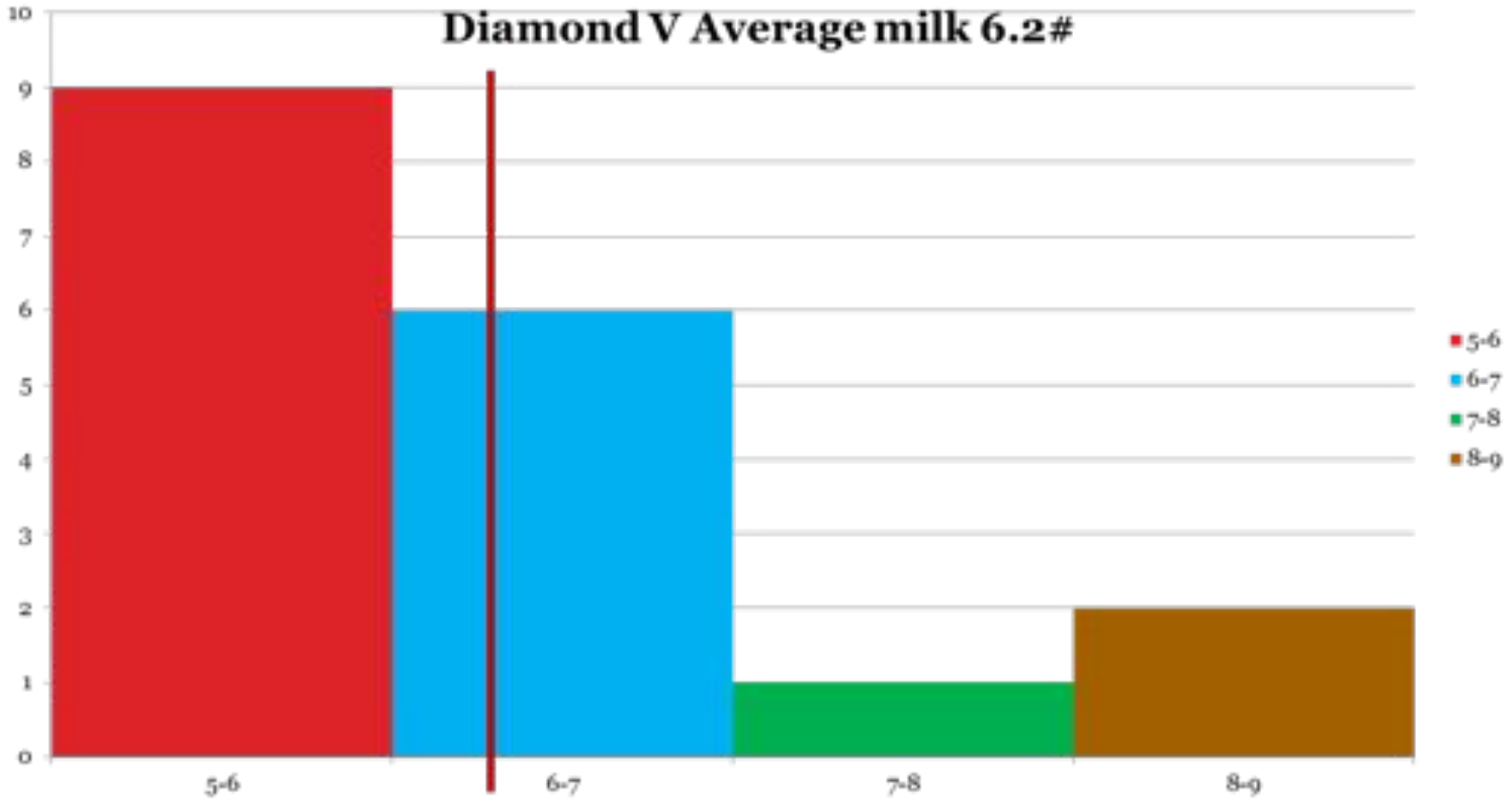
Diamond V Yeast Culture Lactating Dairy Studies
N = 51 Studies, Mean = 3.32 lb FCM



New Diamond V Analysis (only include 10 studies)



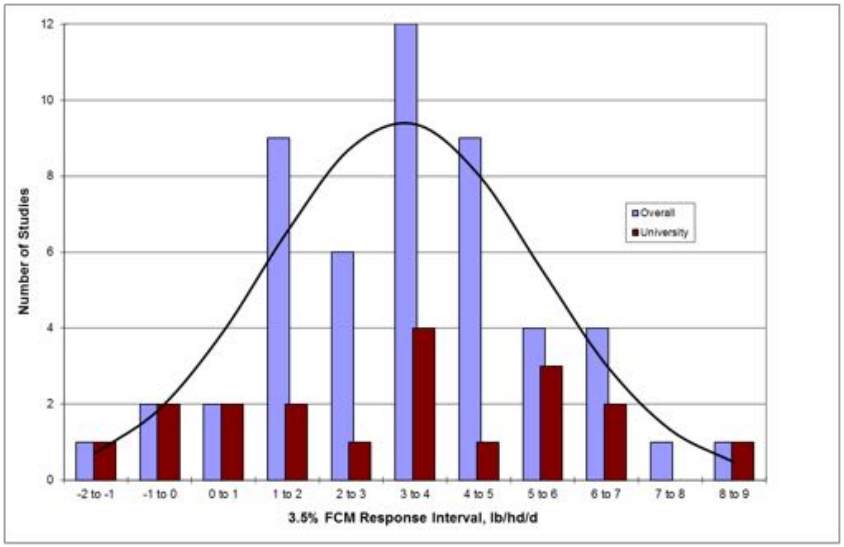
Systematic Review Prevents this

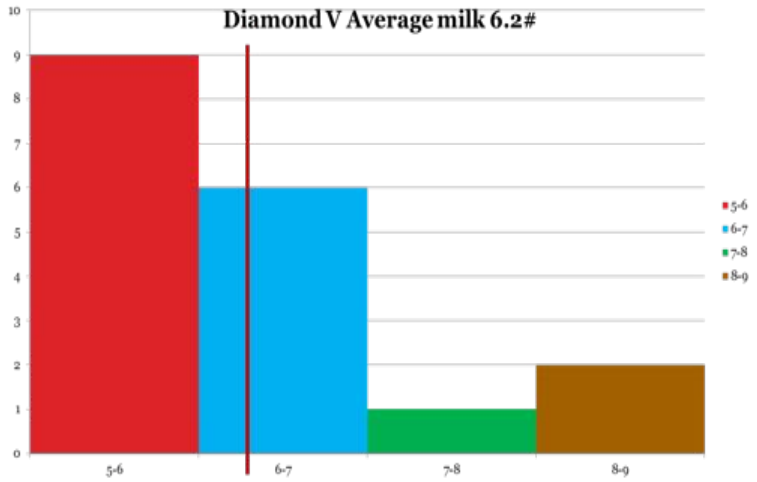


Diamond V

Publication Bias

- Method for looking at if people have been hiding papers. (no bell curve)





Diamond V

One Study

- What do we really know if only one study is done on a product or intervention.
  - We don't know if it is one of the "95%" or of the "5%"
  - With 2 studies we start to estimate the true mean.
  - With 5 studies we start to have confidence, but how do we combine the out-comes?
  - Are all studies created equal?

Diamond V

Meta-Analysis

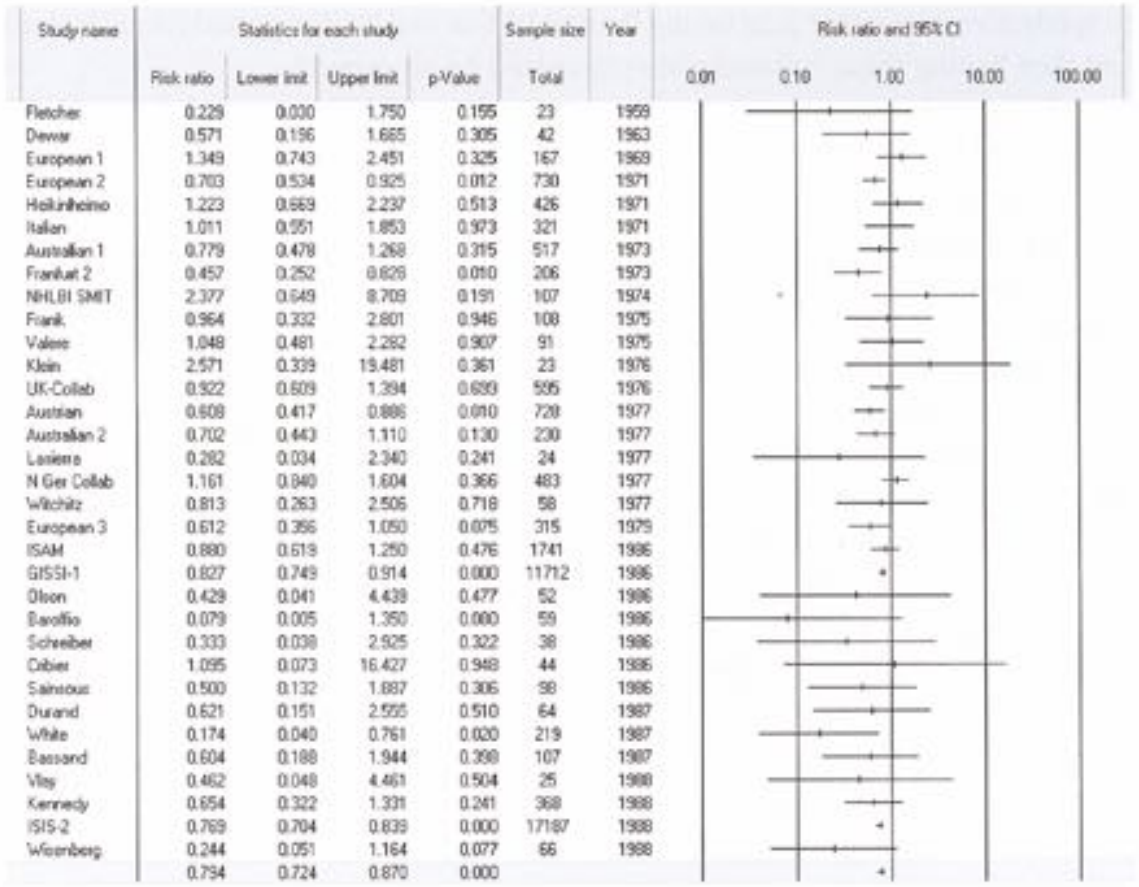
- What is a Meta-Analysis
  - Mathematical Summary of multiple studies
- Why is this important
  - Provides the best evidence from a scientific basis for the risk of using a product or an intervention
    - Example 2x versus 3x milking.
  - The more studies the less uncertainty

Diamond V

Streptokinase example

- Steptokinase is a clot buster that is injected into people following a heart attack
  - Hypothesized to dissolve the clot that is causing the heart attack thus increasing odds of survival
- From 1959 – 1988 there were 33 RCT
  - Different size trials, 23 to 17,187 patients
  - 6 trials showed significant effect
  - 27 showed no significance
  - Would you want streptokinase if you had a heart attack? Only 6/33 with significant effect

Forest Plot of steptokinase mortality



Study name	Statistics for each study				Sample size	Year
	Risk ratio	Lower limit	Upper limit	p-Value	Total	
Fletcher	0.229	0.030	1.750	0.155	23	1959
Dewar	0.571	0.156	1.665	0.305	42	1963
European 1	1.349	0.743	2.451	0.325	167	1969
European 2	0.703	0.534	0.925	0.012	730	1971
Heikinheimo	1.223	0.669	2.237	0.513	426	1971
Italian	1.011	0.551	1.853	0.973	321	1971
Australian 1	0.779	0.478	1.268	0.315	517	1973
Frankat 2	0.457	0.252	0.826	0.010	206	1973
NHLBI SMIT	2.377	0.649	8.709	0.191	107	1974
Frank	0.964	0.332	2.801	0.946	108	1975
Valere	1.048	0.481	2.282	0.907	91	1975
Klein	2.571	0.339	19.481	0.361	23	1976
UK-Collab	0.922	0.609	1.394	0.699	595	1976
Austrian	0.608	0.417	0.886	0.010	728	1977
Australian 2	0.702	0.443	1.110	0.130	230	1977
Lasierra	0.282	0.034	2.340	0.241	24	1977
N Ger Collab	1.161	0.840	1.604	0.366	483	1977
Witchitz	0.813	0.263	2.505	0.718	58	1977
European 3	0.612	0.356	1.050	0.075	315	1979
ISAM	0.880	0.619	1.250	0.476	1741	1986
GISSI-1	0.827	0.749	0.914	0.000	11712	1986
Olson	0.429	0.041	4.439	0.477	52	1986
Barolfio	0.079	0.005	1.350	0.080	59	1986
Schreiber	0.333	0.038	2.925	0.322	38	1986
Cribier	1.095	0.073	16.427	0.948	44	1986
Sainsous	0.500	0.132	1.897	0.306	98	1986
Durand	0.621	0.151	2.555	0.510	64	1987
White	0.174	0.040	0.761	0.020	219	1987
Bassand	0.604	0.188	1.944	0.398	107	1987
Vlay	0.462	0.048	4.461	0.504	25	1988
Kennedy	0.654	0.322	1.331	0.241	368	1988
ISIS-2	0.769	0.704	0.839	0.000	17187	1988
Wisenberg	0.244	0.051	1.164	0.077	66	1988
	0.794	0.724	0.870	0.000		

Figure 2.1 Impact of streptokinase on mortality (adapted from Lau *et al.*, 1992).

- Effect size left of center indicates more likely to survive
  - Effect size is reasonably consistent
  - RR .79 (95% CI 87-92) **--21%** decrease in risk of death
  - *p*= .0000008

Diamond V

Results of Streptokinase

- In this example 6 studies significant, 27 not significant led most to conclude from a narrative review
  - Evidence against an effect
  - Or results were inconsistent
- Meta–analysis
  - Allows us to combine the effects and evaluate the statistical significance of the summary effect. *p*=.0000008
    - 8 chances in 10 million that the drug was not effective.
    - Allowed doctors to save many more lives from heart attacks.

Reducing Uncertainty

- A properly done meta-analysis is the best tool to give us information for reducing uncertainty.

Diamond V

What is a Meta-analysis?

- Is a systematic method that takes data from a number of independent studies, all meeting an inclusion criteria, and integrates them using statistical analysis.

Diamond V

Diamond V Meta-Analysis

J. Dairy Sci. 95:6027–6041
http://dx.doi.org/10.3168/jds.2012-5577

A meta-analysis of the effects of feeding yeast culture produced by anaerobic fermentation of *Saccharomyces cerevisiae* on milk production of lactating dairy cows

G. D. Poppy,*†[1] A. R. Rabiee,‡ I. J. Lean,‡ W. K. Sanchez,† K. L. Dorton,† and P. S. Morley*
*Department of Clinical Sciences, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, Fort Collins 80523
†Diamond V, Cedar Rapids, IA 52405
‡SBEScibus, PO Box 660, Camden 2570, NSW, Australia

ABSTRACT

The purpose of this study was to use meta-analytic methods to estimate the effect of a commercially available yeast culture product on milk production and other production measures in lactating dairy cows using a meta-analysis of randomized controlled trials. Sixty-one research publications (published journal articles, published abstracts, and technical reports) were identified through a review of literature provided by the manufacturer and a search of published literature using 6 search engines. Thirty-six separate studies with 69 comparisons met the criteria for inclusion in the meta-analysis. The fixed-effect meta-analysis showed substantial heterogeneity for milk yield, energy-corrected milk, 3.5% fat-corrected milk, milk fat yield, and milk

findings provide strong evidence that this commercially available yeast culture product provides significant improvement in several important milk production outcomes as evaluated in production settings typical for commercial dairies in North America. Utilizing meta-analytic methods to study the complete breadth of information relating to a specific treatment by studying multiple overcomes of all eligible studies can reduce the uncertainty often seen in small individual studies designed without sufficient power to detect differences in treatments.

Key words: yeast culture, meta-analysis, lactating dairy cow

INTRODUCTION

Poppy et al., J Dairy Sci, 2012

Diamond V

Materials and Methods

- 61 research publications were identified through a review of Diamond V's literature and search of published literature using 6 search engines.

- 36 separate studies with 69 comparisons met the criteria for inclusion.

Diamond V

Large number of Studies

- The more studies, done properly, help us examine and understand the uncertainty or risk around products or interventions
- The information tells us how variable and uncertain the outcomes from the studies are.
- Allows for building a “Risk” Distribution of the economic outcomes.

Diamond V

Materials and Methods

- Inclusion Criteria:
  - Studies must have evaluated at least one Diamond V Original product (YC™,XP™, or XPC™)
  - Studies must have included a negative control group and randomized treatment assignments

Diamond V

Materials and Methods

- Inclusion Criteria (cont.):
  - Studies must have been conducted in lactating cows (not dry cows or in vitro studies)
  - Studies must have used a parallel group design (i.e., not crossover)
  - Studies must have reported results of at least one production outcome: MY, ECM, FY, PY, 3.5% FCM, DMI

Diamond V

Publication bias

Diamond V

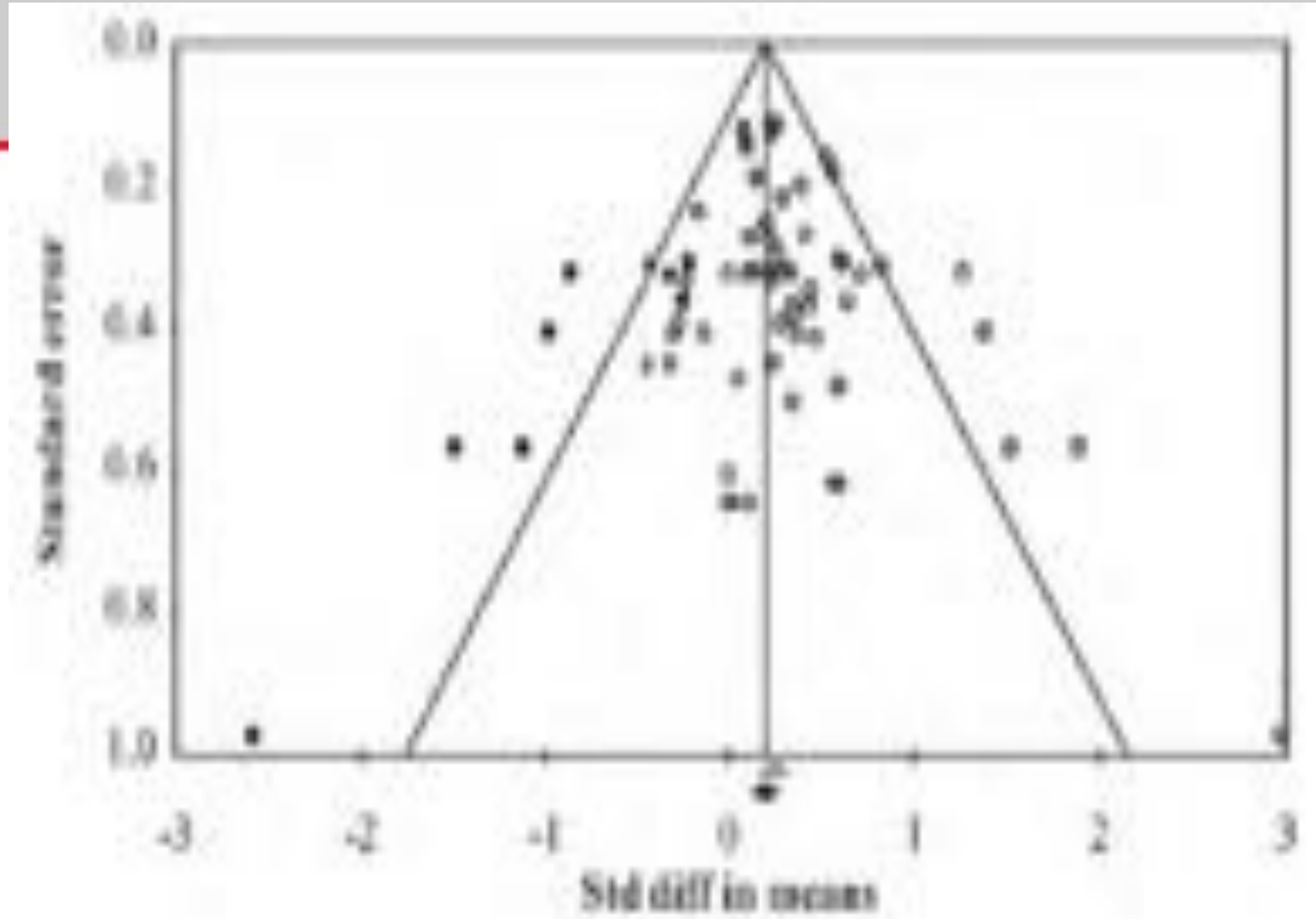
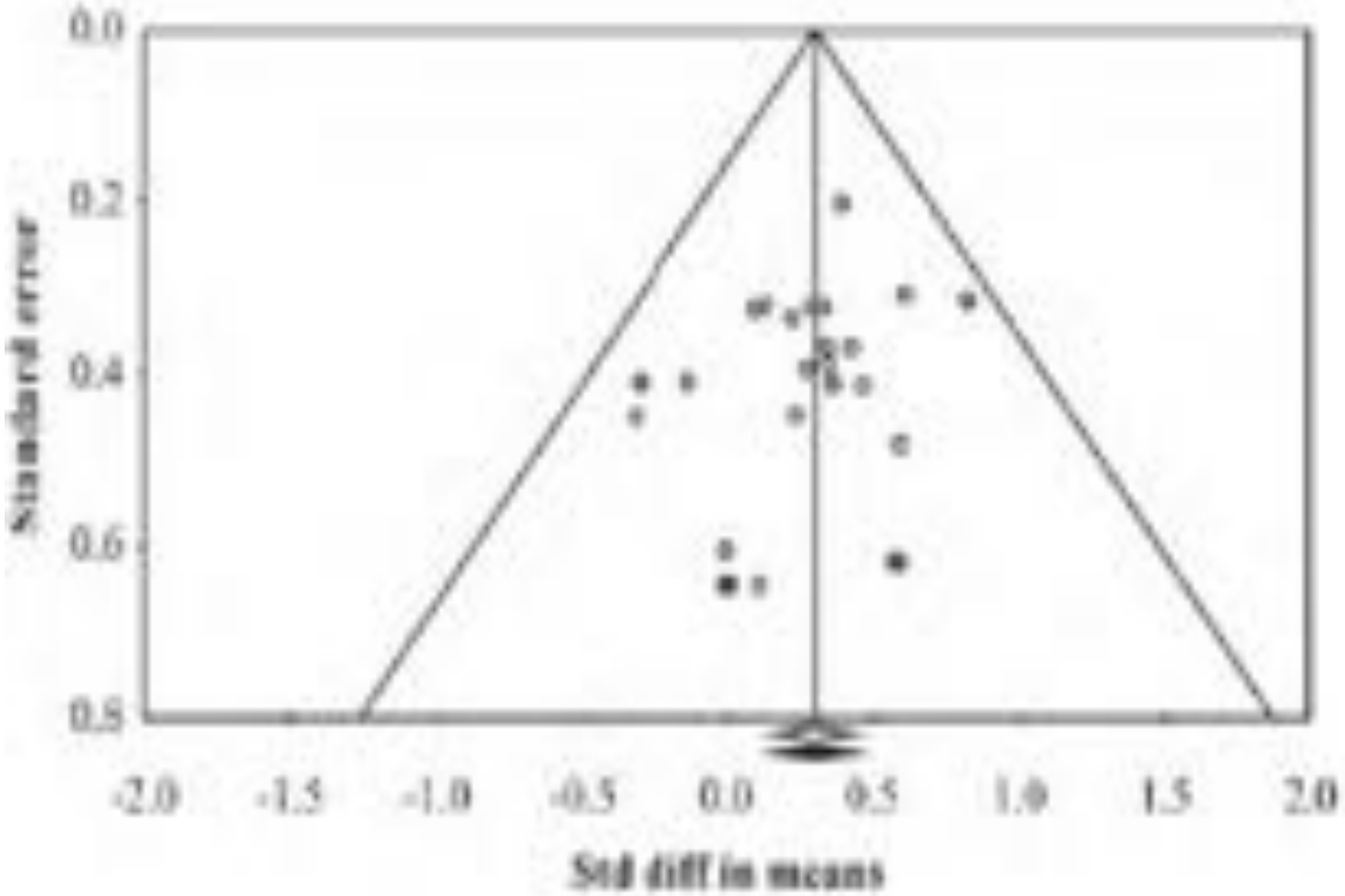


Figure 3. Funnel plot of the standardized mean difference (Std. diff in means) of studies (empty circles) from all studies with milk yield data, meeting the criteria to be included in the meta-analysis

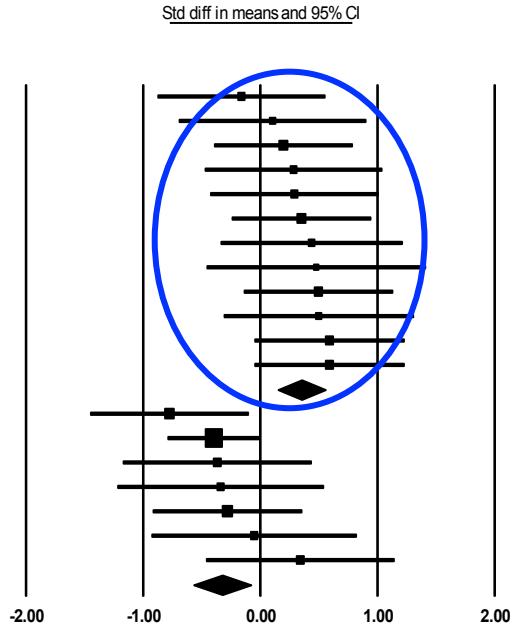
Analysis of Peer Reviewed Only – Milk Production



There were not any missing studies when the Trim and Fill procedure was used. Thus, the two Standardized Mean Differences did not differ.

DMI by Stage of Lactation – Forest Plot

Group by Stage of lactation 2	Study name	Std diff in means	Standard error	Variance	Lower limit	Upper limit	Z-Value	p-Value
Early	Erasmus et al., 2005 - 1	-0.161	0.366	0.134	-0.878	0.556	-0.441	0.659
Early	Wang et al., 2001 - 1- 17% NDF	0.105	0.409	0.167	-0.696	0.906	0.257	0.797
Early	Ramsing et al., 2009 - 4X	0.197	0.302	0.091	-0.396	0.789	0.651	0.515
Early	Robinson, 1997	0.284	0.387	0.150	-0.475	1.042	0.733	0.464
Early	Erasmus et al., 2005 - 2	0.290	0.367	0.135	-0.429	1.010	0.791	0.429
Early	Ramsing et al., 2009 - 1X	0.351	0.304	0.092	-0.244	0.947	1.155	0.248
Early	Robinson - Garrett, 1999 - 2	0.438	0.397	0.158	-0.340	1.216	1.104	0.270
Early	Robinson - Garrett, 1999 - 1	0.477	0.478	0.229	-0.460	1.414	0.999	0.318
Early	Dann et al., 2000 - 3	0.496	0.325	0.106	-0.141	1.134	1.526	0.127
Early	Wang et al., 2001 - 1- 21% NDF	0.499	0.415	0.172	-0.314	1.311	1.203	0.229
Early	Dann et al., 2000 - 2	0.590	0.327	0.107	-0.051	1.232	1.803	0.071
Early	Dann et al., 2000 - 1	0.590	0.327	0.107	-0.051	1.232	1.803	0.071
Early		0.350	0.103	0.011	0.148	0.552	3.394	0.001
Not Early	Harris et al., 1992	-0.776	0.346	0.119	-1.453	-0.098	-2.245	0.025
Not Early	Alshaikh et al., 2002	-0.397	0.202	0.041	-0.793	-0.001	-1.964	0.049
Not Early	Wang et al., 2001 - 2- 17% NDF	-0.367	0.412	0.169	-1.174	0.439	-0.893	0.372
Not Early	Cooke et al., 2007	-0.339	0.450	0.203	-1.222	0.544	-0.753	0.452
Not Early	Schingoethe et al., 2004	-0.280	0.326	0.106	-0.919	0.359	-0.859	0.390
Not Early	Arambel and Kent, 1990	-0.053	0.447	0.200	-0.929	0.824	-0.118	0.906
Not Early	Wang et al., 2001 - 2- 21% NDF	0.341	0.411	0.169	-0.465	1.147	0.830	0.407
Not Early		-0.327	0.125	0.016	-0.572	-0.082	-2.617	0.009



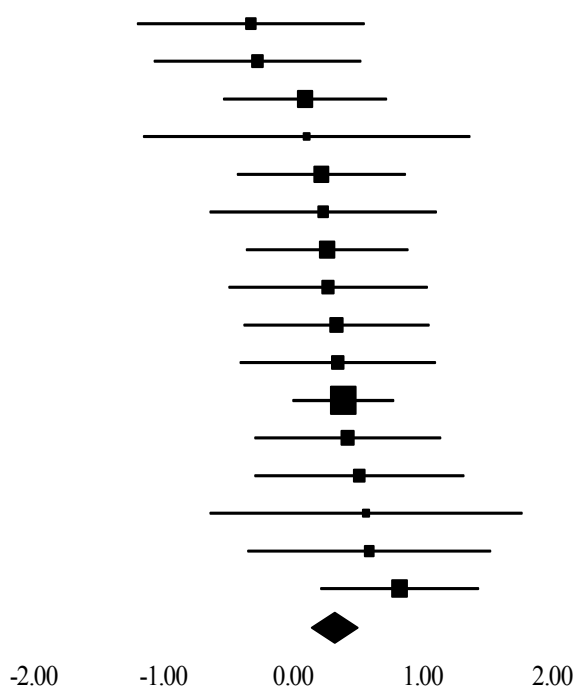
Meta Analysis

Poppy et al., J Dairy Sci, 2012

Diamond V

Forest Plot for Milk Yield

Arambel and Kent, 1990	-0.316	0.450	-1.198	0.566	0.482	10	10
Wang et al., 2001 - Syn 17%NDF	-0.265	0.410	-1.069	0.538	0.518	12	12
Schingoethe et al., 2004	0.101	0.325	-0.535	0.738	0.755	19	19
Lehloenya et al., 2008 - Primi	0.114	0.646	-1.152	1.380	0.860	4	6
Harris et al., 1992	0.227	0.334	-0.428	0.883	0.497	18	18
Cooke et al., 2007	0.241	0.449	-0.639	1.121	0.591	10	10
Dann et al., 2000 -Syn	0.272	0.322	-0.359	0.903	0.398	19	20
Robinson - Garrett, 1999 - 2	0.279	0.394	-0.493	1.052	0.479	13	13
Erasmus et al., 2005 - 1	0.344	0.368	-0.377	1.065	0.350	15	15
Robinson, 1997	0.354	0.388	-0.407	1.115	0.362	13	14
Alshaikh et al., 2002	0.396	0.202	0.001	0.792	0.050	50	50
Erasmus et al., 2005 - 2	0.430	0.369	-0.294	1.154	0.244	15	15
Wang et al., 2001 - Syn 21%NDF	0.520	0.415	-0.294	1.333	0.210	12	12
Lehloenya et al., 2008 -Multi	0.572	0.618	-0.638	1.783	0.354	6	5
Robinson - Garrett, 1999 - 1	0.596	0.482	-0.348	1.540	0.216	9	9
Ramsing et al., 2009 - Syn	0.831	0.314	0.215	1.447	0.008	22	22
	0.321	0.091	0.143	0.499	0.000		



Scientific Validation

CHANGE IN DMI AND MILK YIELD*

	Difference in DMI			Difference in Milk Yield		
Stage of Lactation	**lb/ head/ day**	**P-value**	**95% CI**	**lb/ head/ day**	**P-value**	**95% CI**
First 70 days of lactation	1.37	0.003	0.46 to 2.24	3.02	0.001	1.21 to 3.98
Mid-late lactation	-1.72	0.008	-2.99 to -0.46	2.16	0.049	0.02 to 4.29

* Poppy et al., 2012, J. Dairy Sci.

Diamond V

Overall Results – Peer-reviewed studies

Item, Lbs.	Response	95% CI	P- value
3.5% FCM	3.55	2.0 – 5.1	0.001
ECM	3.64	2.1 – 5.2	0.001
Fat	0.13	0.02 - 0.22	0.009
Protein	0.07	0.0 – 0.11	0.026

* Poppy et al., 2012, J. Dairy Sci.

Diamond V

Economics of Response by Stage of Lactation

Item	Early Lactation	Mid-late Lactation
Milk (lb.)	3.01	2.16
Dry Matter Intake (lb.)	+1.36	-1.72
Milk value @ $15/cwt	$0.45	$.32
DMI value @ $.12/lb. DM	($0.16)	$0.20
Diamond V product cost	$0.05	$0.05
Bottom line IOFC (dollars/cow/day)	$0.24	$0.47

Diamond V

Understanding the Numbers

		Mean	SE	Variance	.ower Limi	Jpper Limi	P Value		
Milk Yield	14	3.02	0.83	0.31	1.39	4.65	0.001		

- The SE is the standard error, how sure are we of the mean or average
- The square root of the variance is how much the cows within the herds vary in individual response.

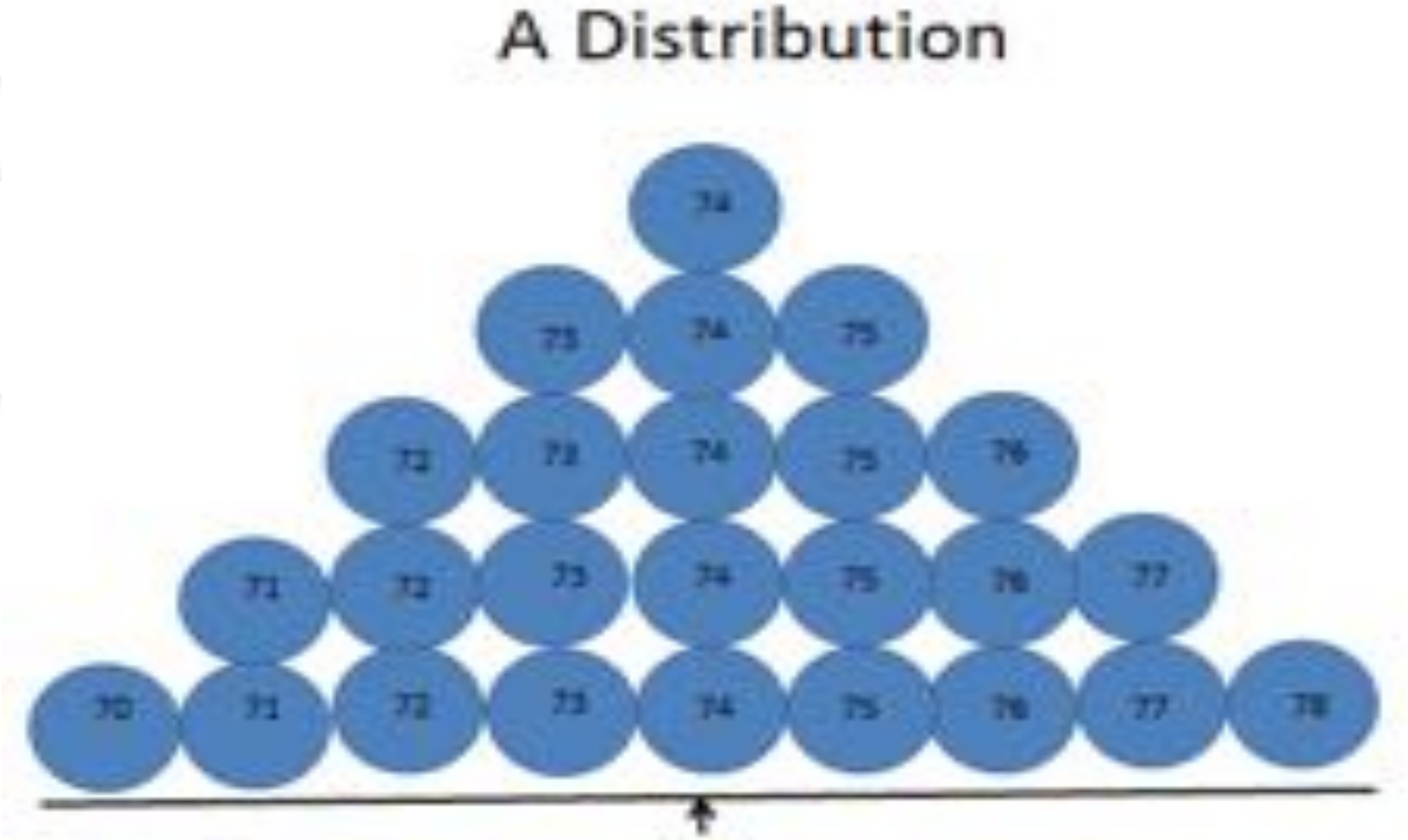
Stochastic Analysis

- Equation - Milk Production x # of Cows x milk Price = Income
- Milk production is 74 but we don't really know what it will be tomorrow.
- The Standard Error tells us it varies from 70 to 78.

Imagine a bin of numbers

- We can look at the distribution of milk production like a bin of numbers.
- The computer reaches into the bin and selects a number randomly weighted by the distribution.

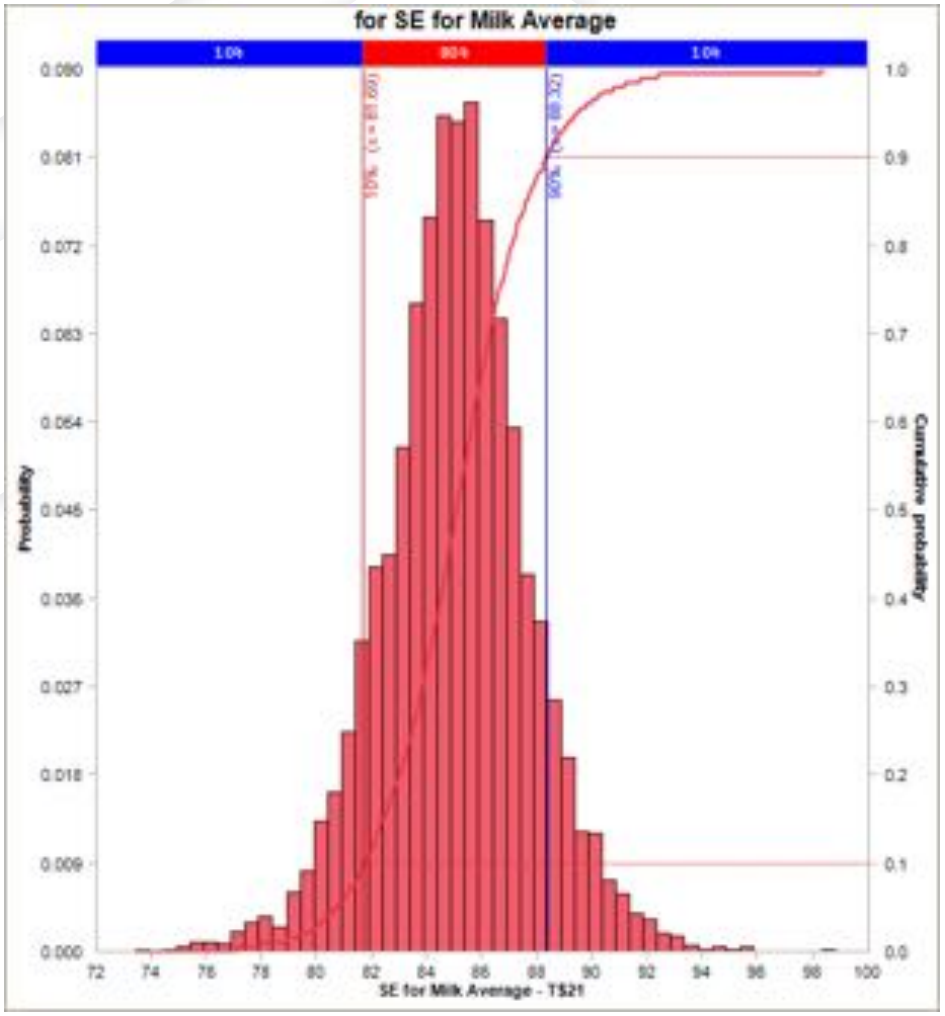
Distribution of balls



Equation

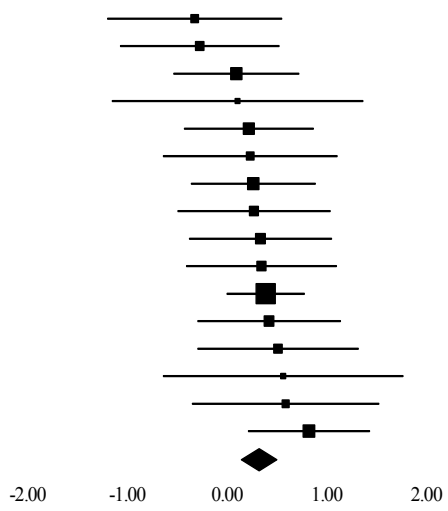
- The computer uses the number to enter into and solve the equation.
- The computer repeats this >5000 times and the answers create a density curve.

The “bin” Milk in Meta-analysis based on the SE



Forest Plot for Milk Yield

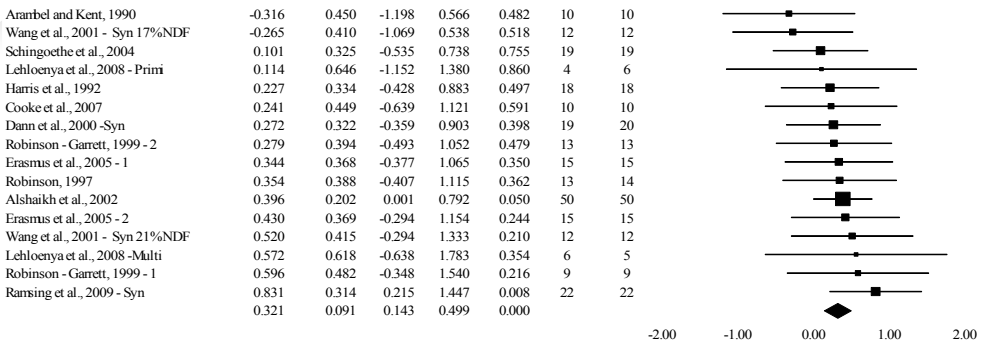
Aranbel and Kent, 1990	-0.316	0.450	-1.198	0.566	0.482	10	10
Wang et al., 2001 - Syn 17%NDF	-0.265	0.410	-1.069	0.538	0.518	12	12
Schingoethe et al., 2004	0.101	0.325	-0.535	0.738	0.755	19	19
Lehloenya et al., 2008 - Primi	0.114	0.646	-1.152	1.380	0.860	4	6
Harris et al., 1992	0.227	0.334	-0.428	0.883	0.497	18	18
Cooke et al., 2007	0.241	0.449	-0.639	1.121	0.591	10	10
Dann et al., 2000 -Syn	0.272	0.322	-0.359	0.903	0.398	19	20
Robinson - Garrett, 1999 - 2	0.279	0.394	-0.493	1.052	0.479	13	13
Erasmus et al., 2005 - 1	0.344	0.368	-0.377	1.065	0.350	15	15
Robinson, 1997	0.354	0.388	-0.407	1.115	0.362	13	14
Alshaikh et al., 2002	0.396	0.202	0.001	0.792	0.050	50	50
Erasmus et al., 2005 - 2	0.430	0.369	-0.294	1.154	0.244	15	15
Wang et al., 2001 - Syn 21%NDF	0.520	0.415	-0.294	1.333	0.210	12	12
Lehloenya et al., 2008 -Multi	0.572	0.618	-0.638	1.783	0.354	6	5
Robinson - Garrett, 1999 - 1	0.596	0.482	-0.348	1.540	0.216	9	9
Ramsing et al., 2009 - Syn	0.831	0.314	0.215	1.447	0.008	22	22
	0.321	0.091	0.143	0.499	0.000		



The bars show Standard Deviation

What are all the sources of variability?

- From the forest plot you can see there is error around the mean – Standard Error
- And uncertainty based on the Standard Deviation of the study.



What is the question?

- We really want to know what is the marginal value of putting an intervention, (DV yeast culture) into the ration and what is the risk of that decision.

The total equation

- (# fluid milk* fluid milk price)+(# BF* BF price) +(# PY * PY price) + (change in DMI * price of DM)
- Each of these components have a both a variance of the mean and a variance for the future outcome that have to be added together.

Sample of Risk Model

- Multiply all the SE and the SD distributions together into a combined distribution for
  - Milk Yield
  - Fat Yield
  - Protein Yield
  - Dry Matter Intake
- Outcome shows the risk of simultaneously analyzing all the risk.

Values for the calculation

- SE and SD are used from the meta-analysis for Milk Yield, Butter Fat, Protein Yield, and DMI.
- Then these values are used to calculate Energy Corrected Milk and a class three milk price is added.
- Milk Price used is $18/cwt in this example.
- DMI cost used is $0.15 /lb

Diamond V ROI calculator

Early Lactation

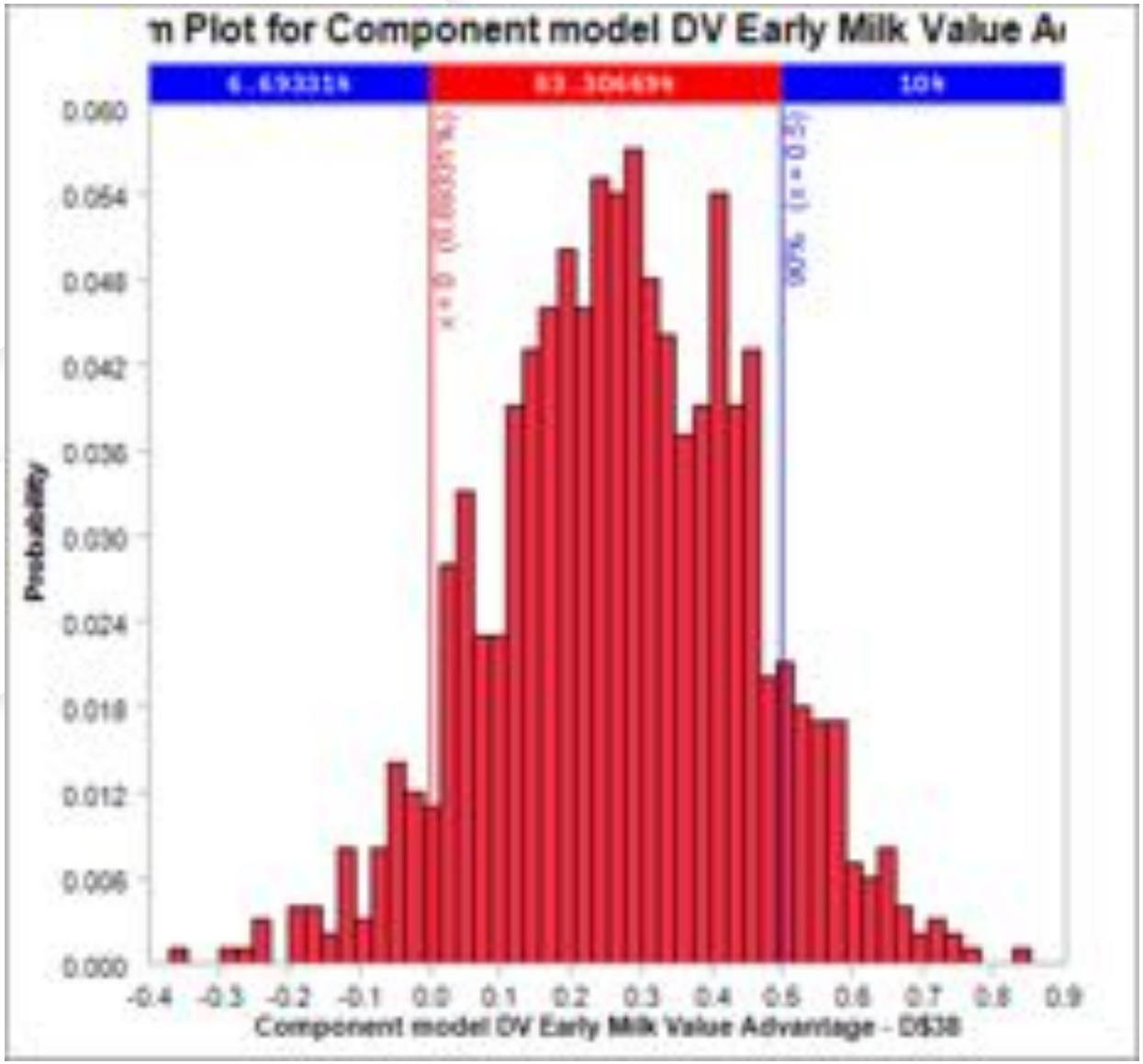
	Control	DV Advantage
Milk Yield, lb	85	88.02
Fat %	3.40%	3.43%
lb of fat	2.89	3.02
Protein %	3.00%	2.96%
lb of Protein	2.55	2.61
OS %	5.70%	5.70%
lb of OS	4.845	5.02
DMI, lb	52	53.36
$ / DM lb	$0.15	
DM, $/cow/	$7.54	$7.74
DV/Cost/Cow		$0.05
$ / cow / day	$16.66	$17.18
$ / cwt	$19.60	$19.52
IOFC Advanta	$9.12	$9.39
DV Milk Value Advantage		$0.27

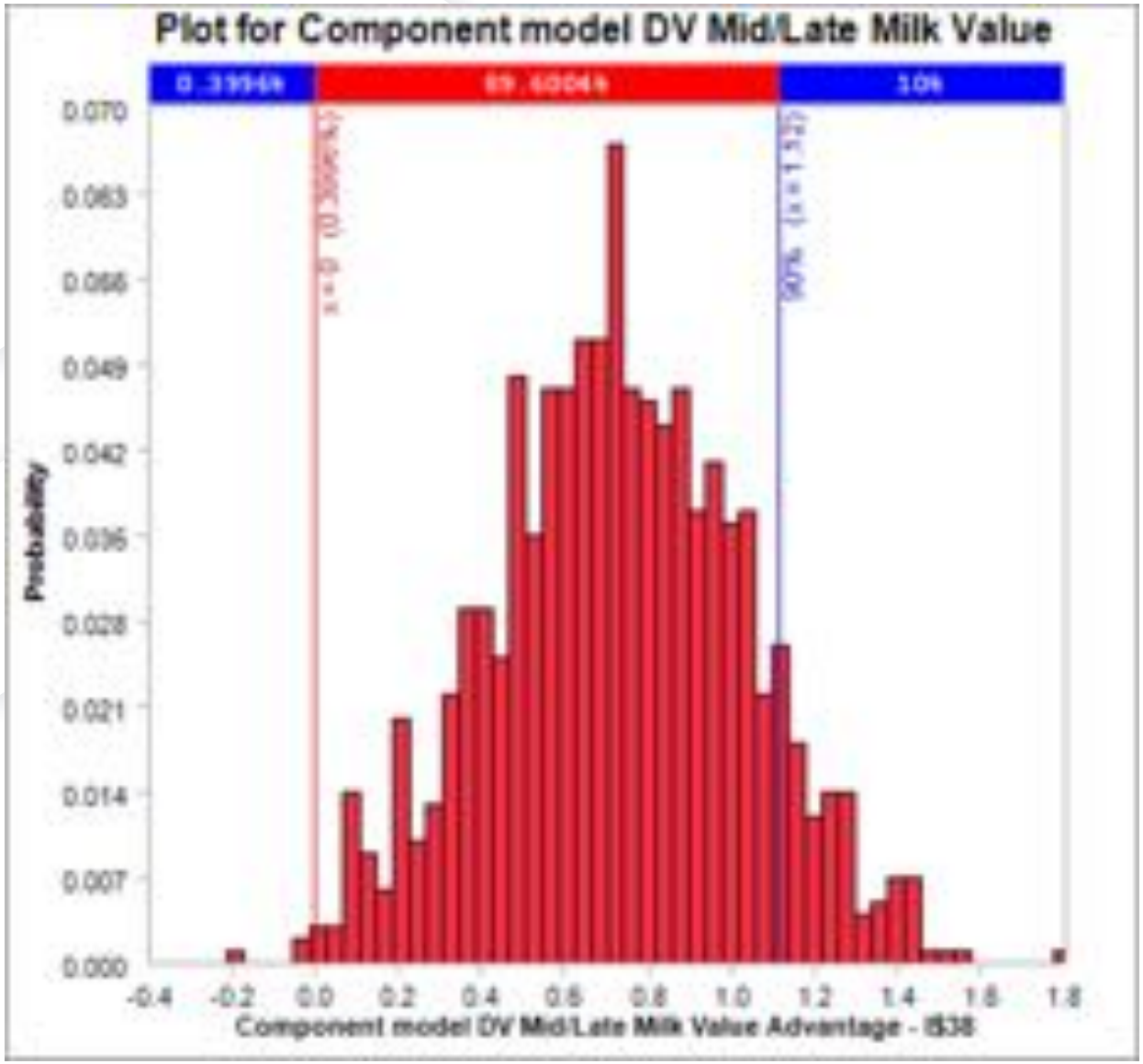
Mid/Late Lactation

	Control	DV Advantage
Milk Yield, lb	70	72.16
Fat %	3.70%	3.74%
Fat, lb	2.59	2.7
Protein %	3.15%	3.18%
Protein, lb	2.205	2.29
OS %	5.70%	5.70%
OS, lb	3.99	4.11
DMI, lb	52	50.28
$ / DM lb	$0.13	
DM, $/cow/c	$6.76	$6.54
DV/Cost/Cow		$0.05
$ / cow / day	$14.41	$14.96
$ / cwt	$20.58	$20.74
IOFC Advanta	$7.65	$8.38
DV Milk Advantage		$0.73

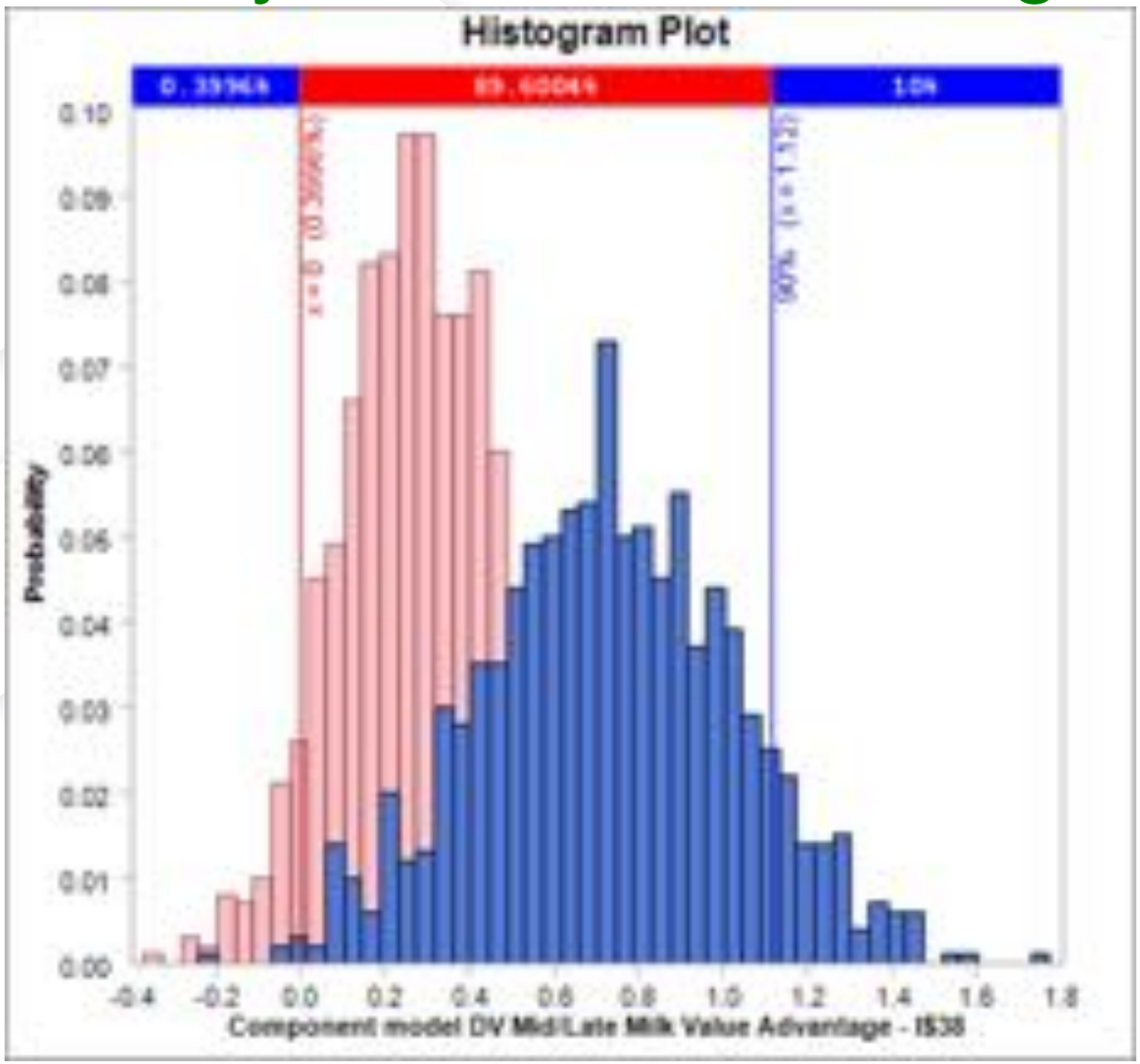
Deterministic Model

- This model is an example of a deterministic model.
- This is how most models are built.
- Once the SD and SE are added they create the density graphs

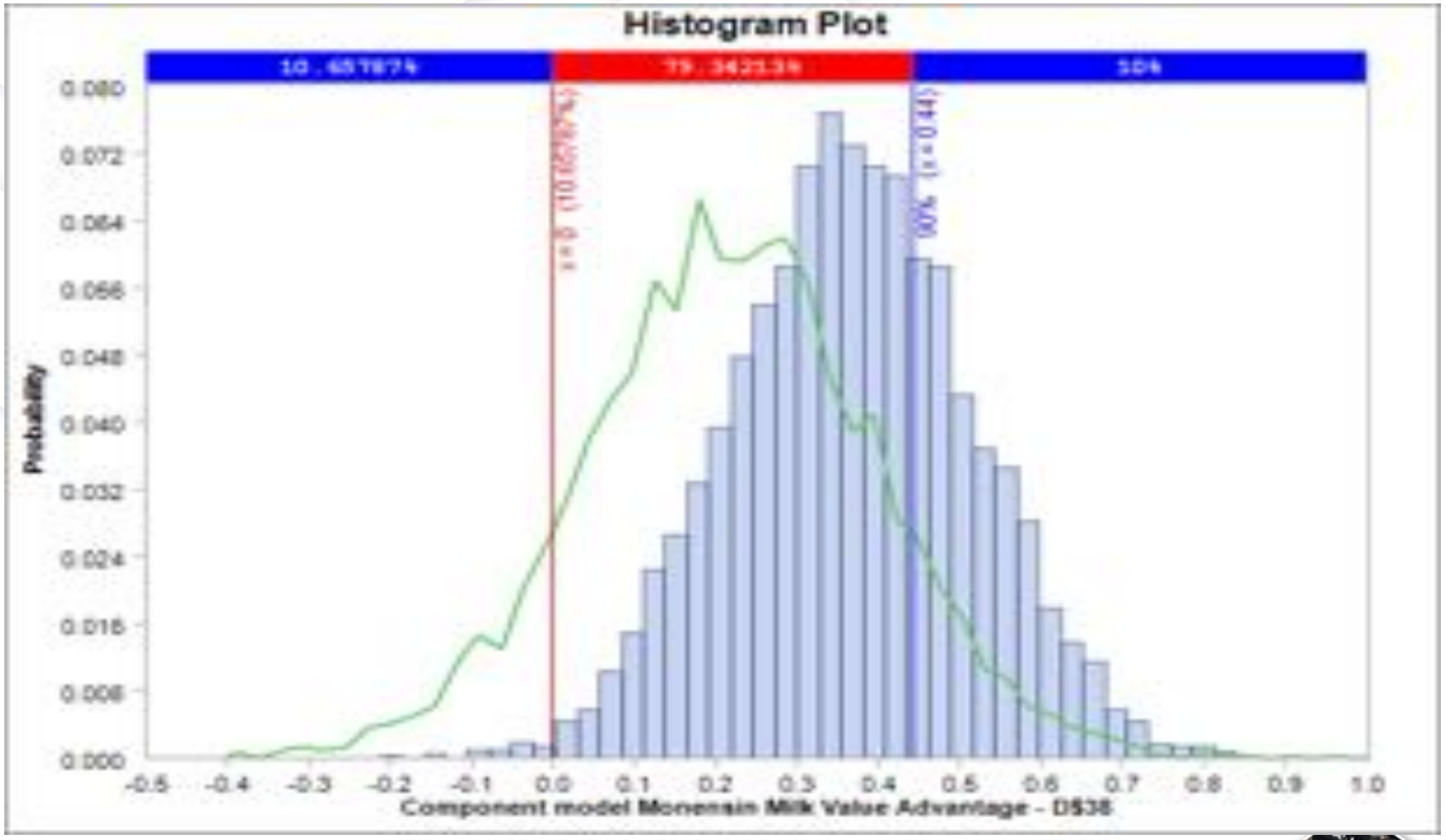

n Plot for Component model DV Early Milk Value A
Probability
0.060
0.054
0.048
0.042
0.036
0.030
0.024
0.018
0.012
0.006
0.000
-0.4 -0.3 -0.2 -0.1 0.0 0.1 0.2 0.3 0.4 0.5 0.6 0.7 0.8 0.9
Component model DV Early Milk Value Advantage - D$38


Plot for Component model DV Mid/Late Milk Value
0.070
0.063
0.056
0.049
0.042
0.035
0.028
0.021
0.014
0.007
0.000
Probability
-0.4
-0.2
0.0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
1.6
1.8
Component model DV Mid/Late Milk Value Advantage - IS38

Both Early and Mid-late histograms



Monensin vs. Diamond V risk



Summary

- Show me the data!
- Ideally, a properly done meta-analysis with Forest plots and Funnel plots
- Large scale, well done field trials are also useful

Diamond V®

The Trusted Experts In Nutrition & Health®