

Handling and Use of Glycerin in Feed

Joe Harris, Ph.D.

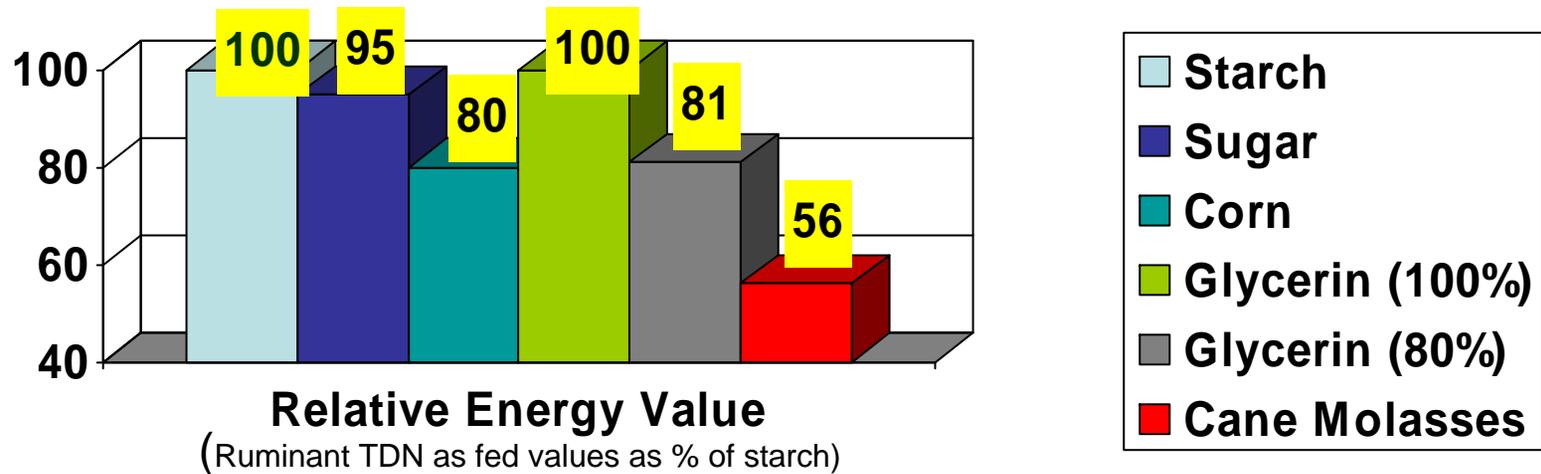
Westway Feed Products, Inc.

Jan. 9, 2007

Objectives of Discussion

- Explain why feed manufacturers have interest in bio-diesel derived glycerin
- Describe how glycerin impacts the physical characteristics, nutritional properties and palatability of feeds
- Describe the developmental process we have used in evaluation of this material as a possible component of our products in terms of:
 - Approved supplier process/ Characterization of available products/ Evaluation of possible contaminants
 - Development of realistic specifications as an ingredient
 - Development of Standard operating procedures for this material
 - Internal evaluation of safety of various sources for use in feed

Why Glycerin?



Why Now?

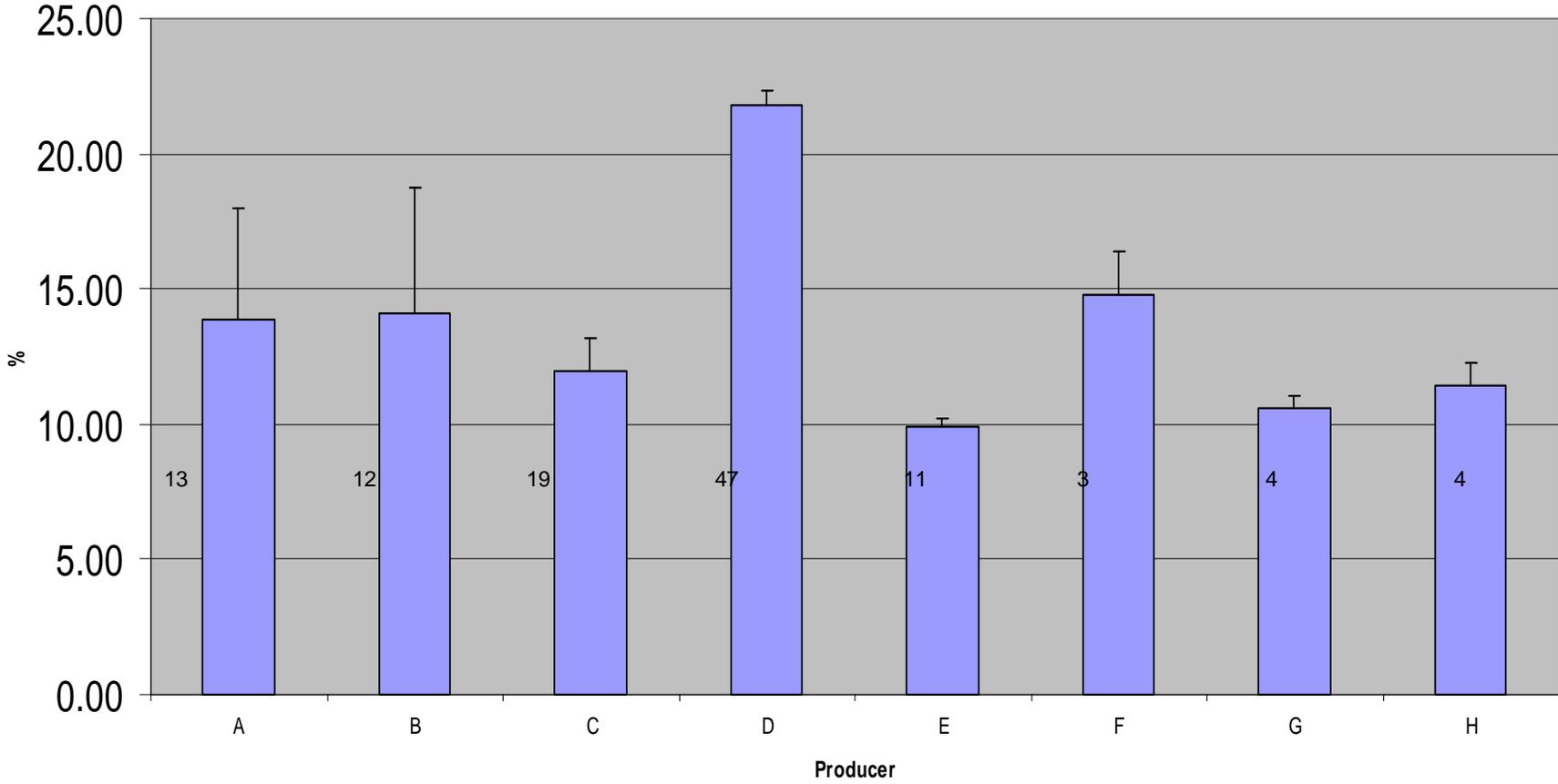
Supplies of glycerin have grown dramatically with increases in bio-diesel production. This has saturated saturating historic uses. Until such time as additional applications are developed-glycerin will compete with traditional energy feeds on a price basis.

Business synergies exists between feed industry and bio-diesel industries

Why Glycerin?

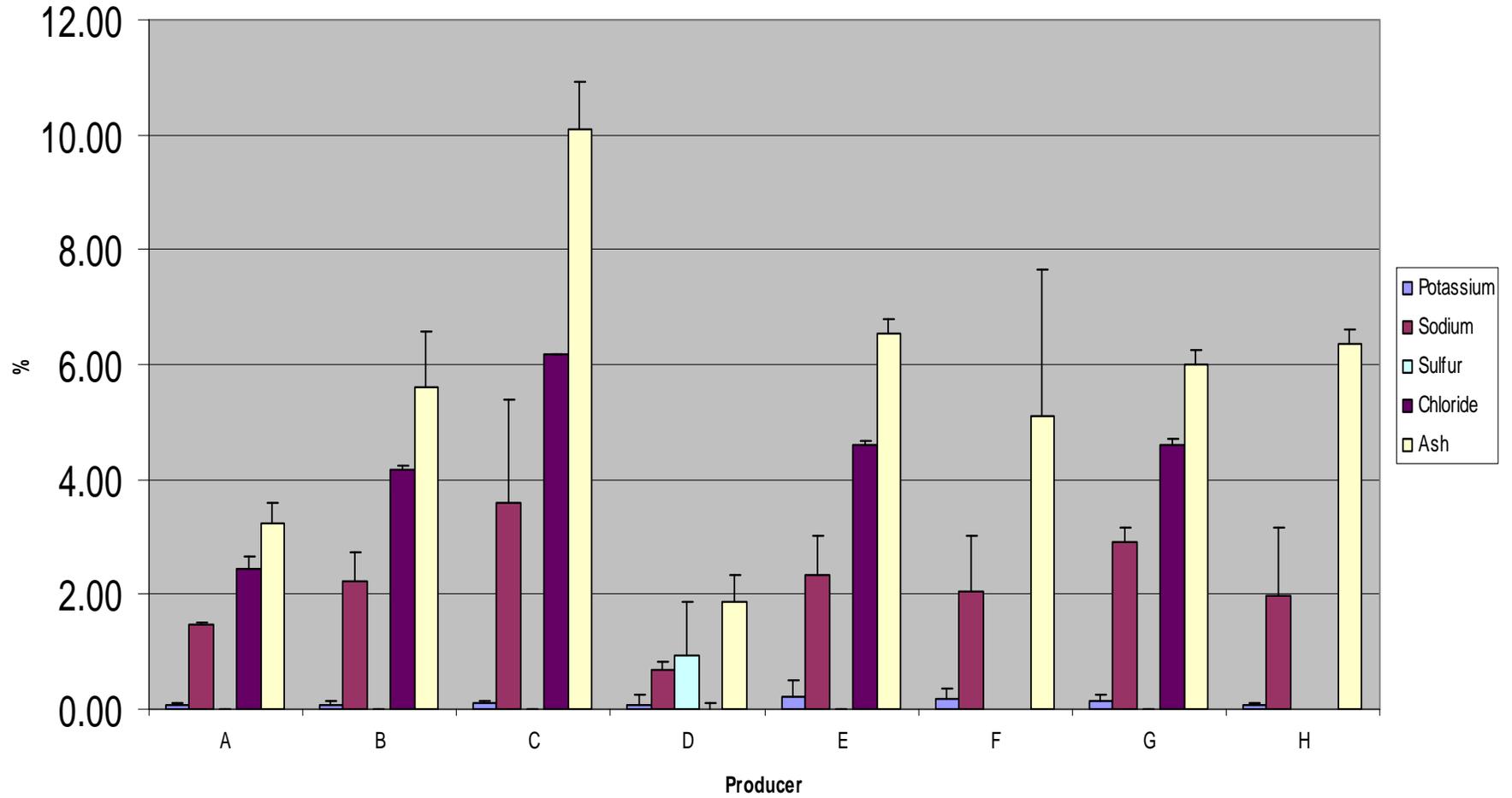
- Glycerin's unique physical properties that make it attractive as a component of liquid feed supplements:
 - Highly water soluble.
 - Melting point of 64 degrees F if absolutely pure but when small amounts of water are present it remains fluid at temperatures near zero.
 - Mild pleasant aroma
 - Sweet Taste
 - Near Neutral ph- typical 5 to 7
 - Highly palatable
 - Decreases the viscosity of molasses and other liquid by-products
 - Hydroscopic-attracts moisture-which can help prevent feeds from “drying out” at low humidity.

Moisture Content of Crude Glycerin By Producer

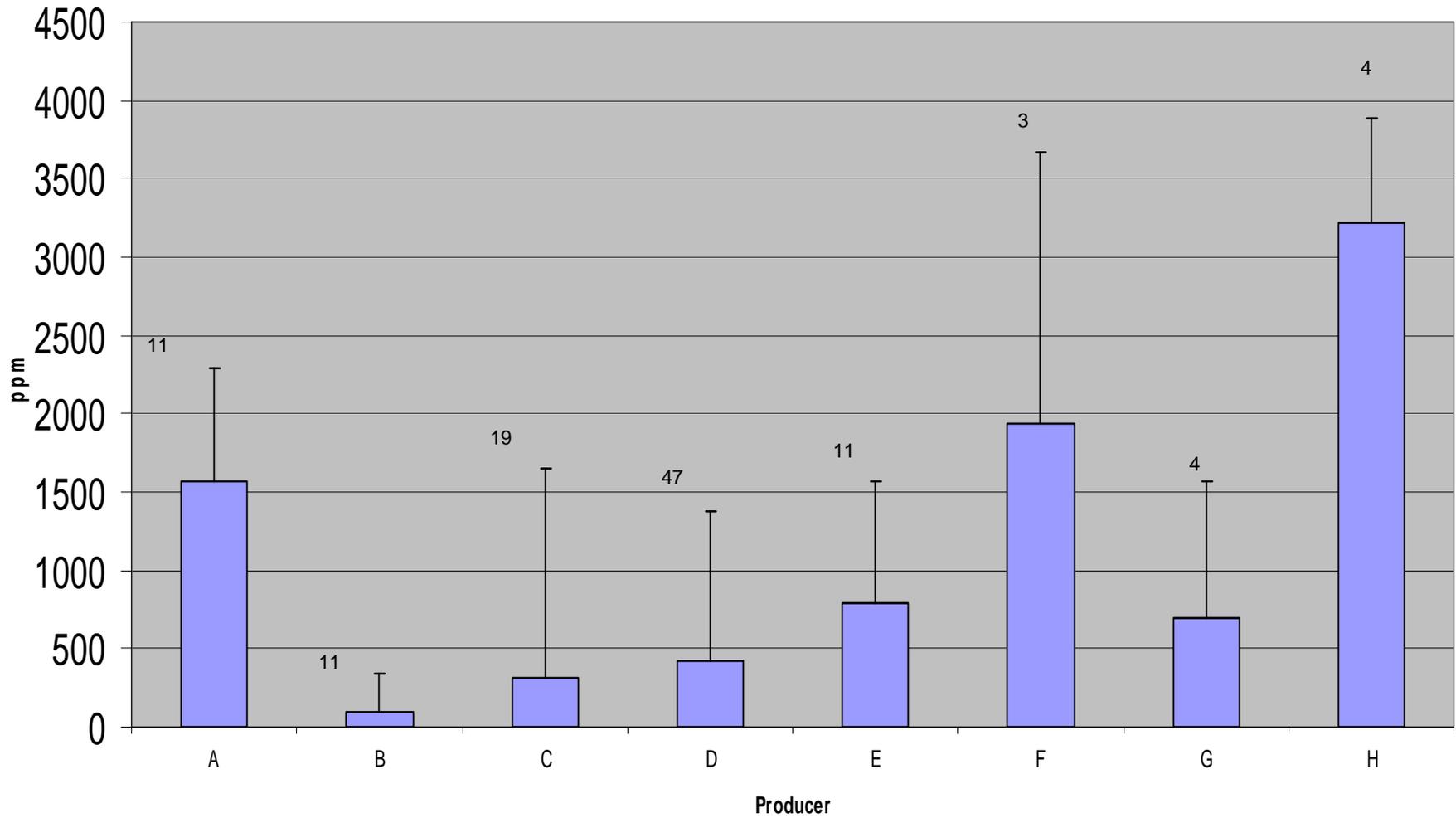


Values shown in bars represent number of samples analyzed

Primary Ash Constituents in Crude Glycerin from Different Producers

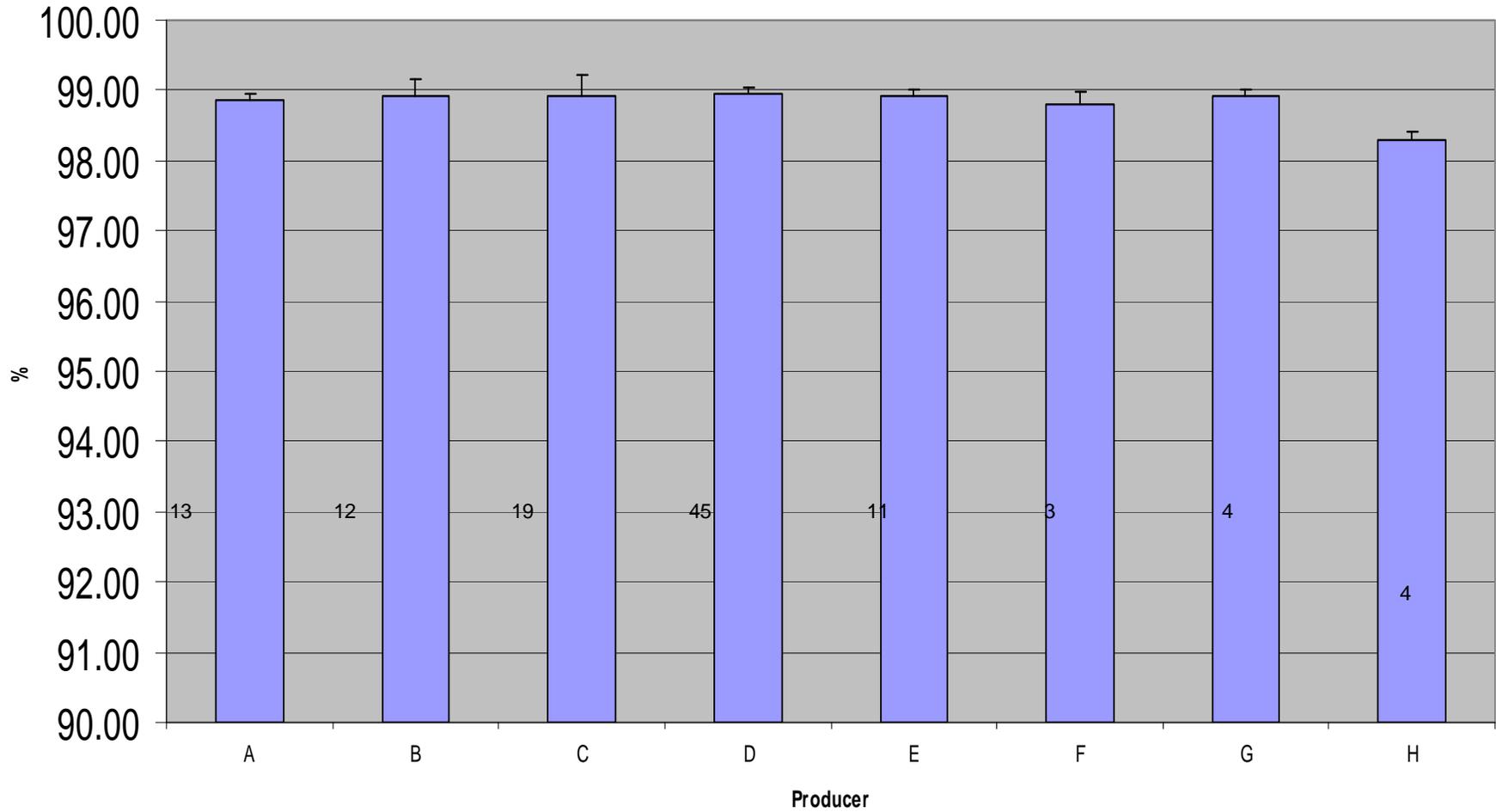


Methanol Content in Crude Glycerin by Producer



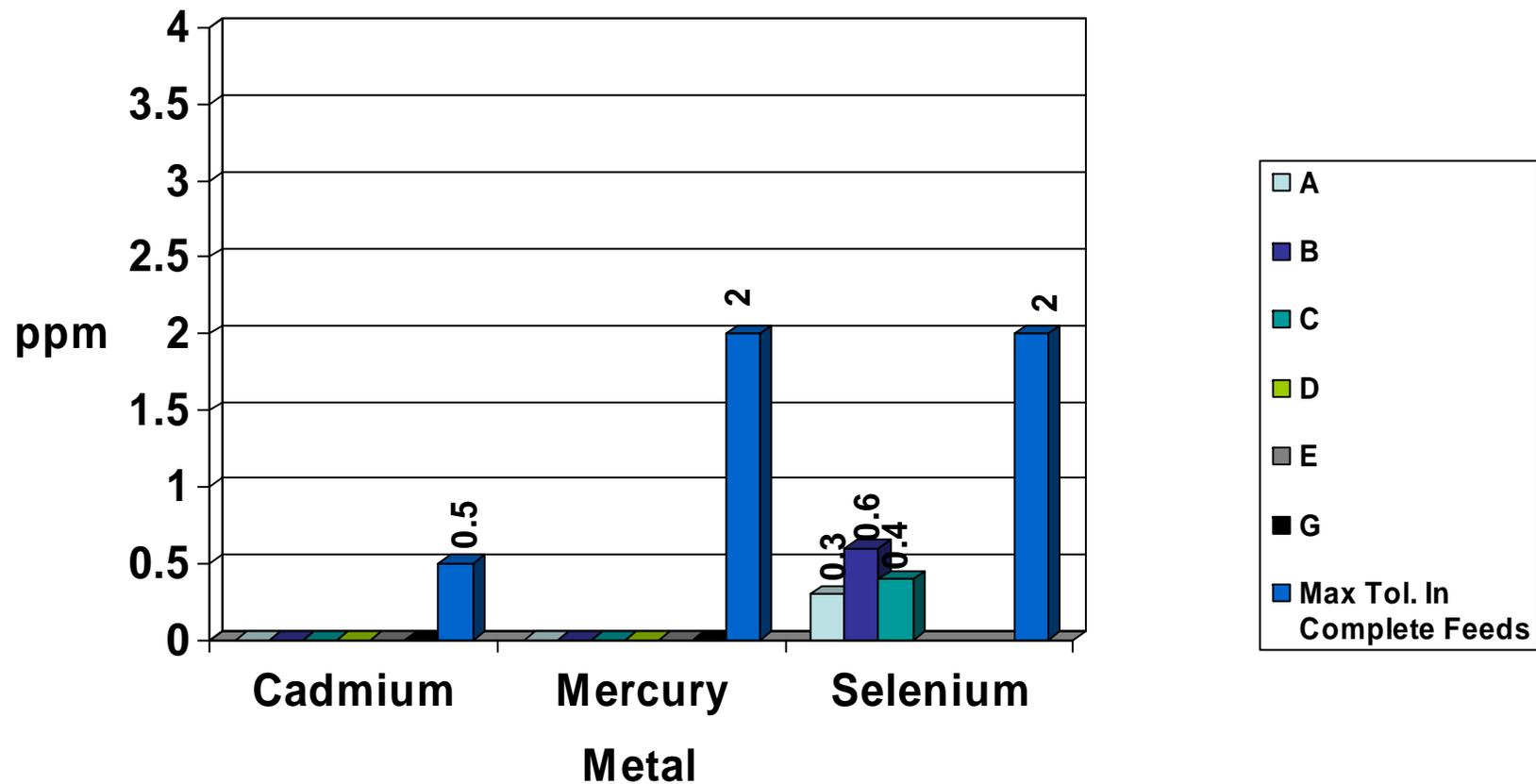
Values shown in bars represent number of samples analyzed

Glycerin Content in Crude Glycerin as a % of Organic Matter by Producer

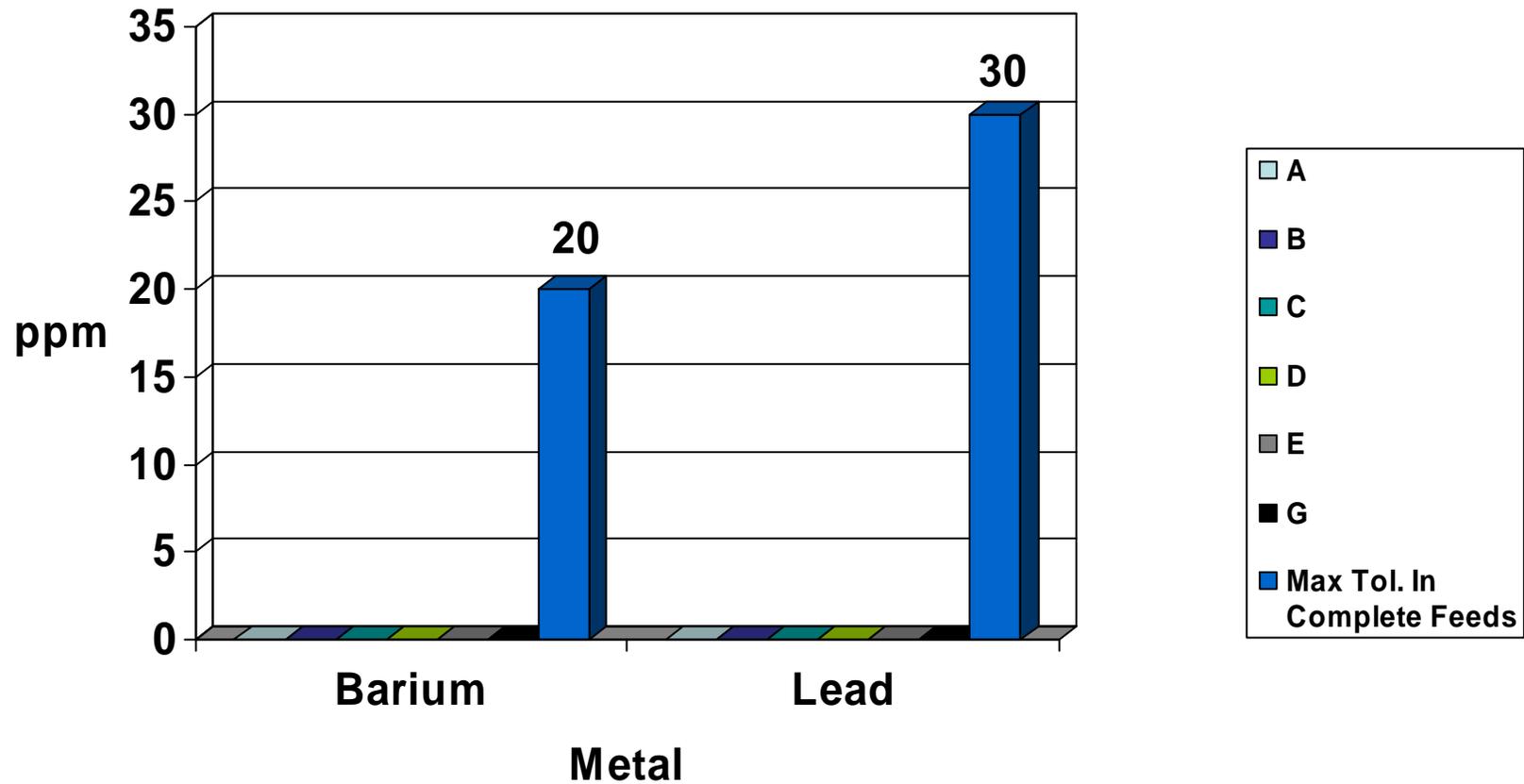


Values shown in bars represent number of samples analyzed

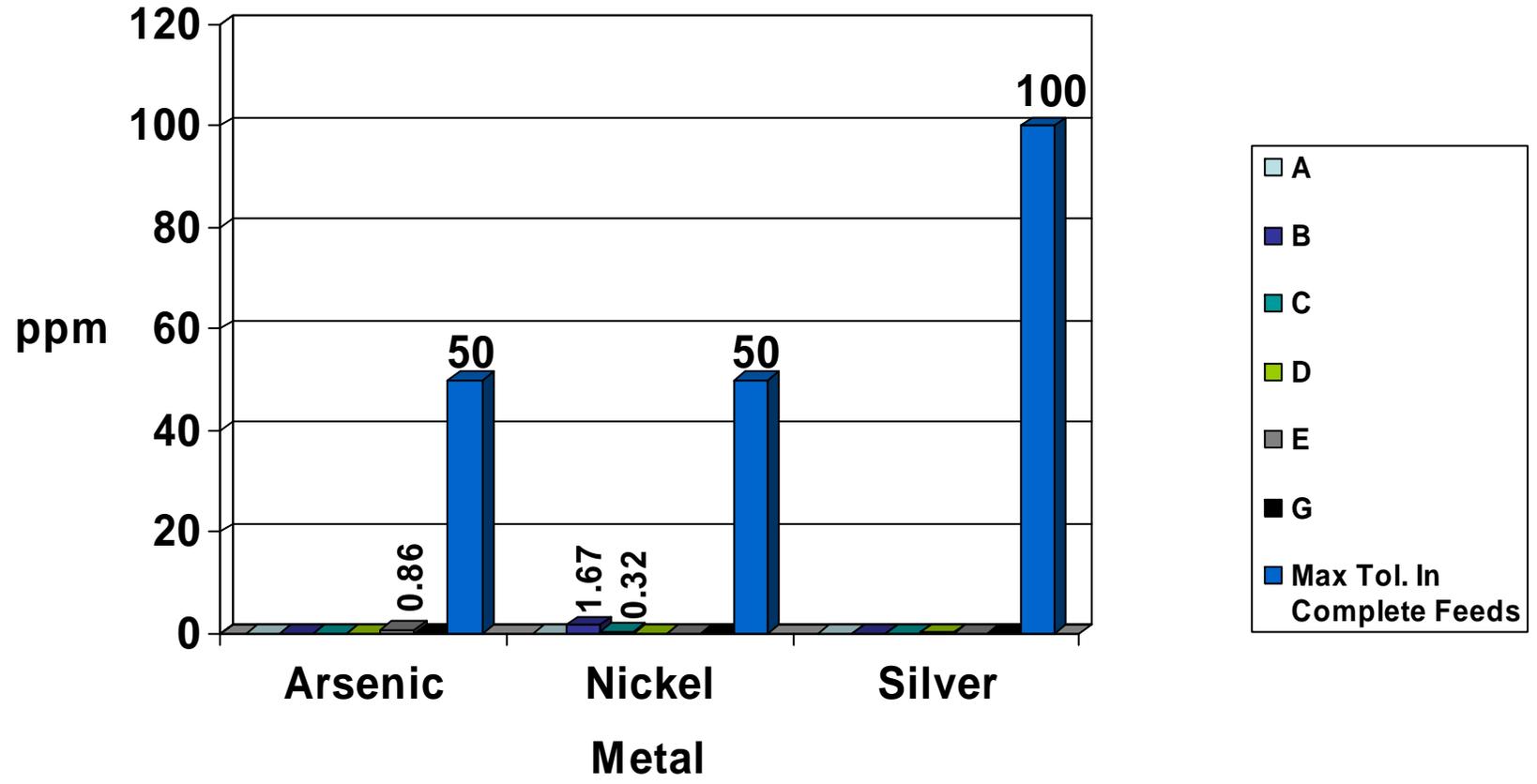
Highly Toxic Heavy Metals in Crude Glycerin



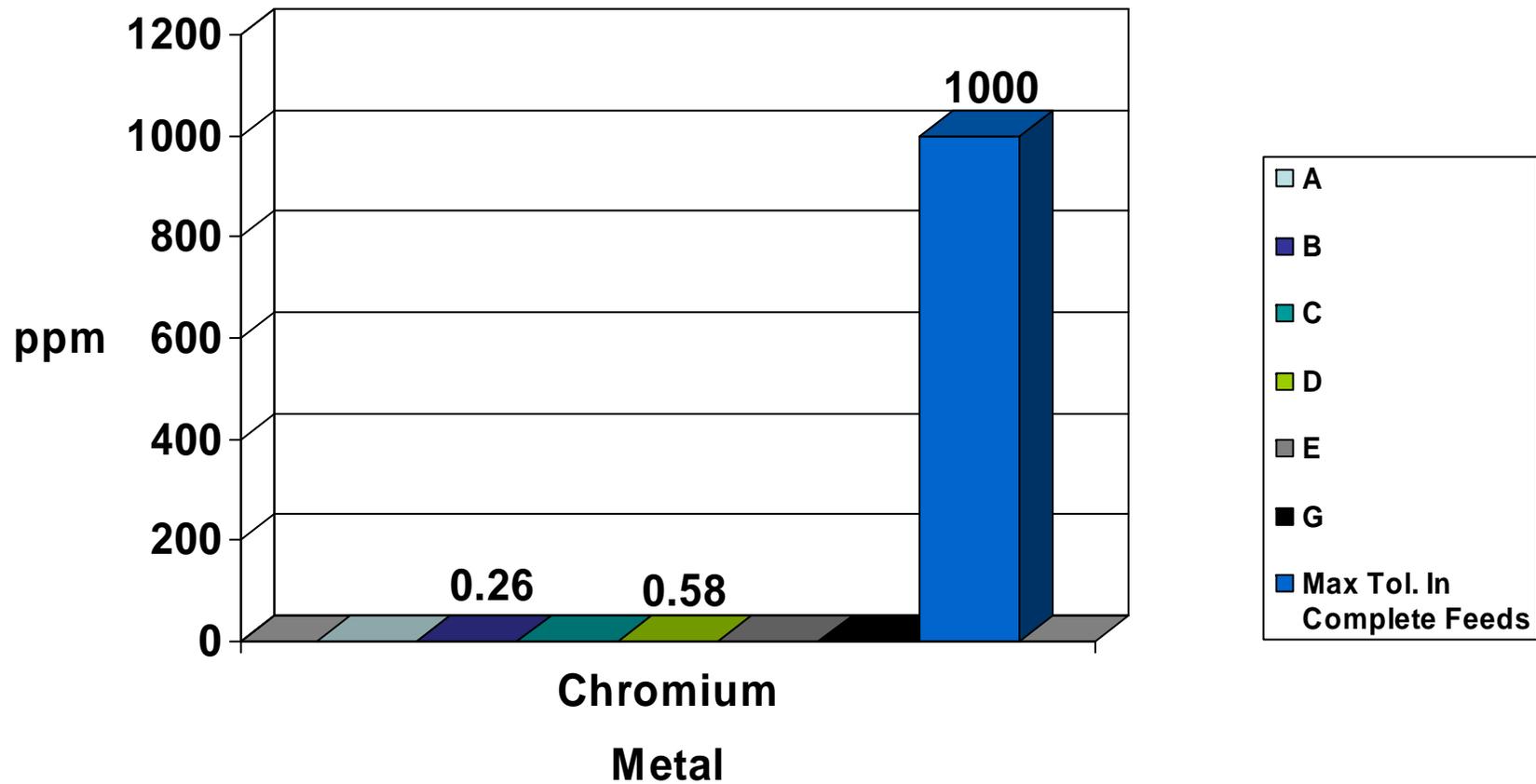
Toxic Heavy Metals in Crude Glycerin



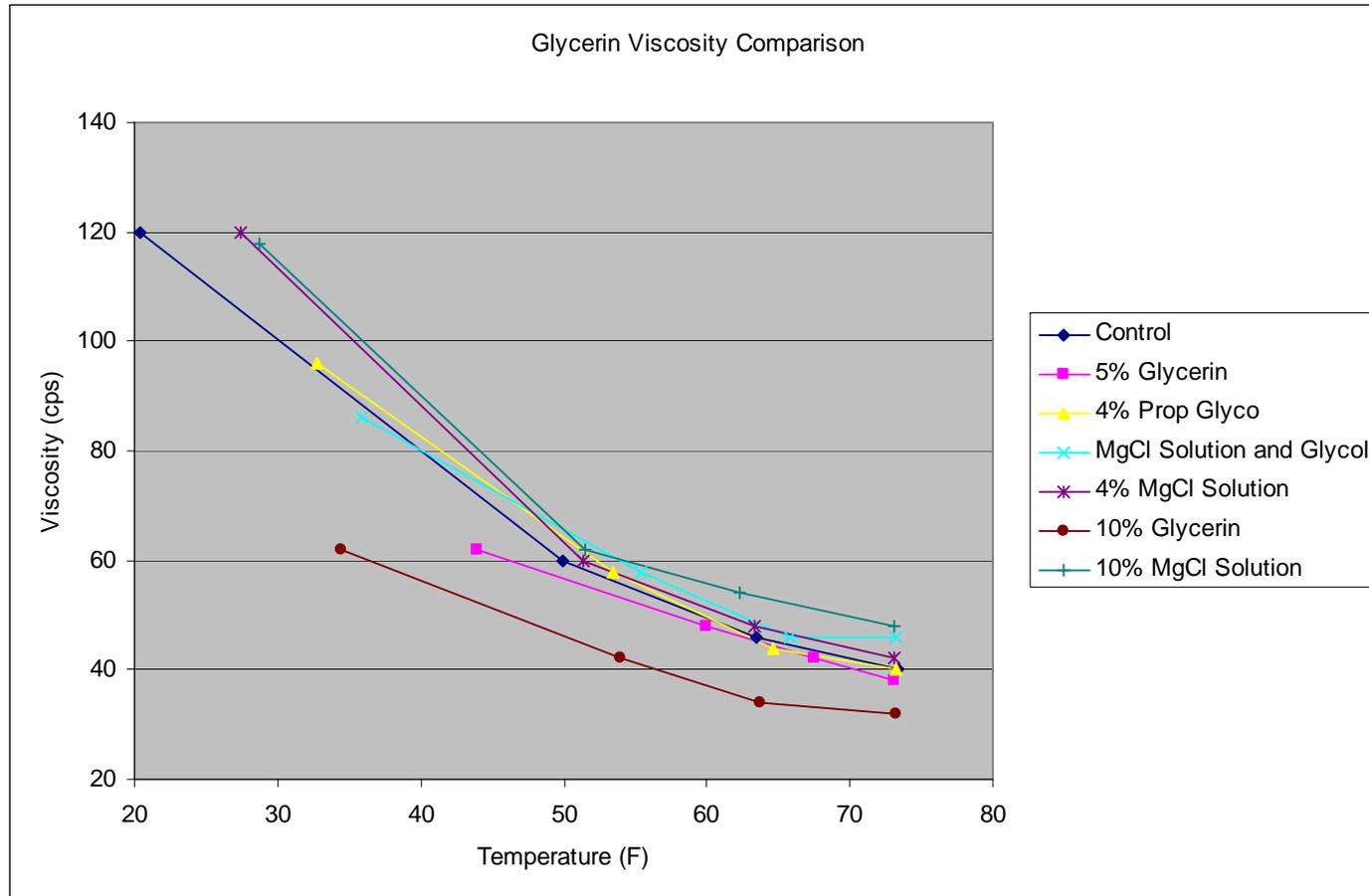
Moderately Toxic Heavy Metals in Crude Glycerin



Slightly Toxic Heavy Metals in Crude Glycerin



Impact of Glycerin on Physical Properties of Textured Feed Conditioner



Evaluation of Liquid Supplements Added to Low Quality Forage in Continuous Culture of Rumen Microbes '06 West Virginia University

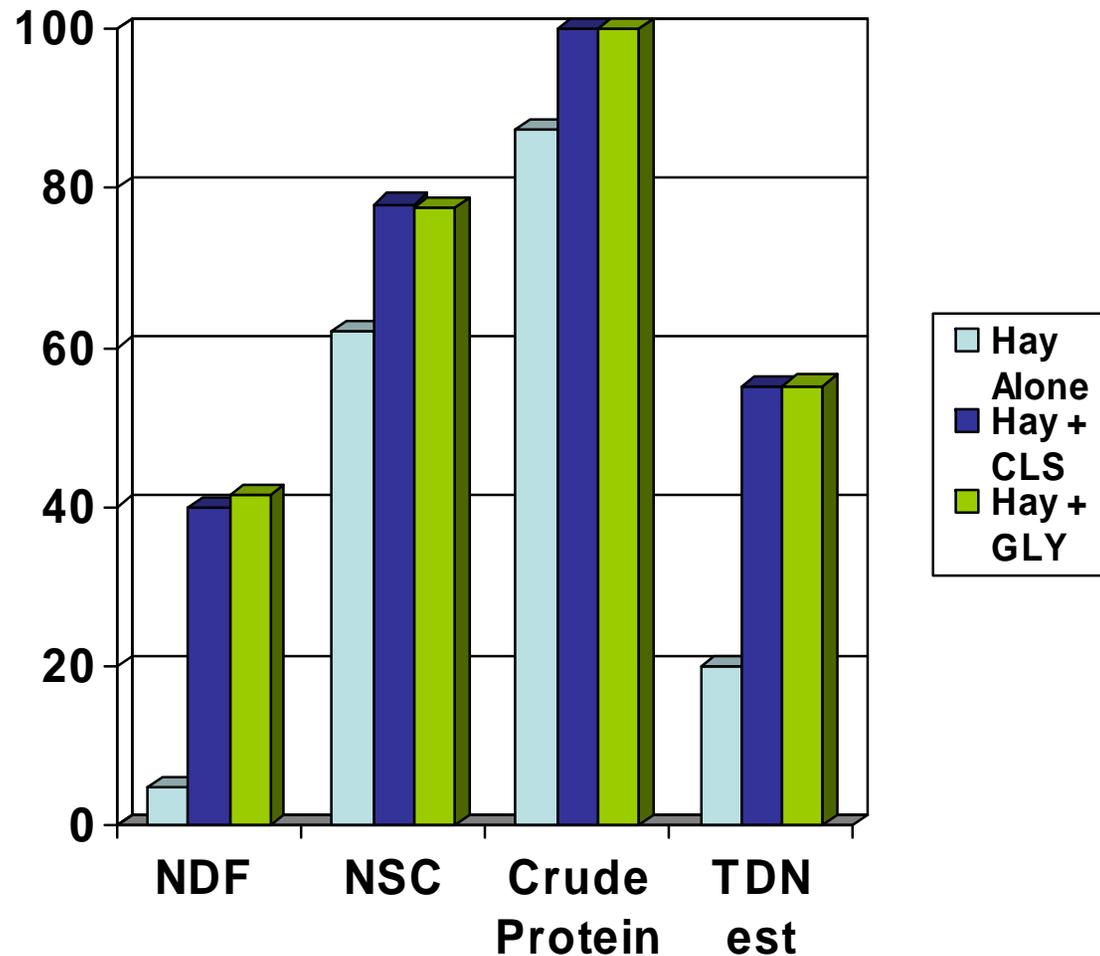
- Procedures
 - A poor quality hay was selected for the study so as to compare well to dormant season grazing pasture conditions. Hay contained 6.3% crude protein and 72.6% Neutral Detergent Fiber
 - Comparisons Included Hay alone vs. Conventional Liquid supplement –exclusively Molasses based (CLS), and Conventional Liquid Supplement containing Molasses + Glycerol (GLY)
 - Liquid supplements were added at 10 % Dry Basis to the Hay in treated cases in an in-vitro continuous culture system.

Evaluation of Liquid Supplements Added to Low Quality Forage in Continuous Culture of Rumen Microbes

Digestibility of Dietary Nutrients %

Results

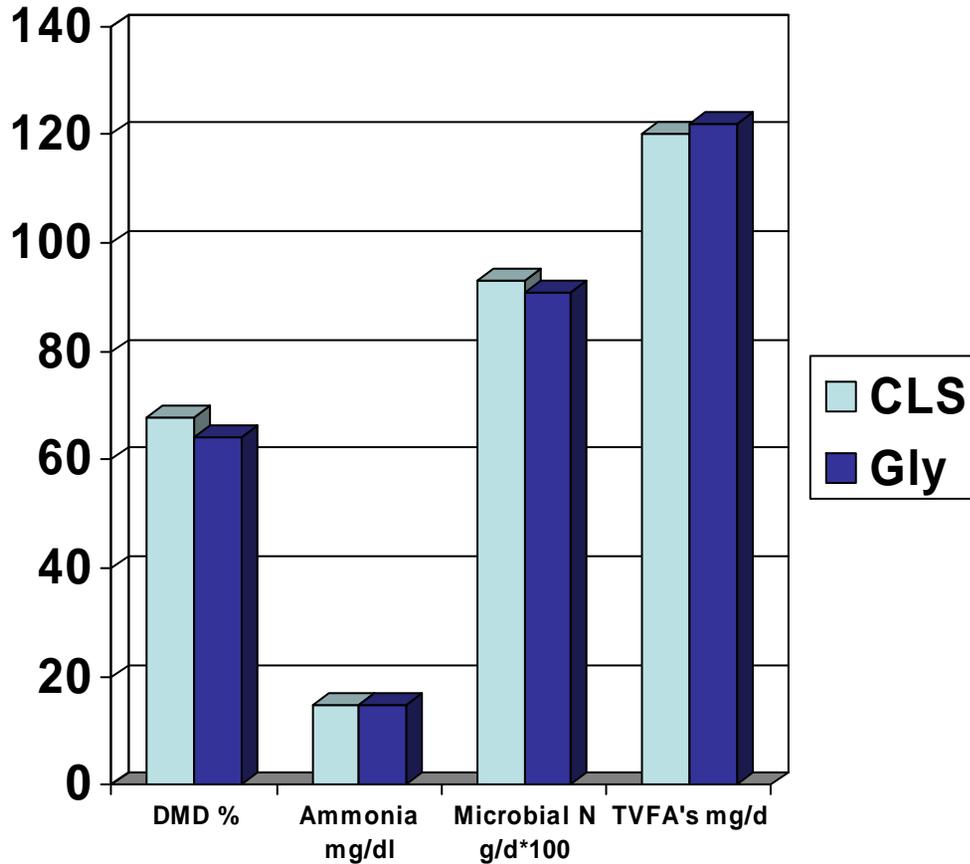
- Dramatic impact on digestibility of dietary fiber (Neutral Detergent Fiber) was observed –Increased by a factor of nearly 8 times
- Digestibility of Non-Structural Carbohydrates (NSC) was high in all cases as expected
- Note TDN est = sum of digestible NDF, NSC and CP



Impact on Measured Parameters

Evaluation of Liquid Supplements (Conventional and 40% Glycerol) Added to Medium Quality Forage

TK Miller, RFPL- WVA10-06



- Study was designed to compare impact replacing molasses with glycerin on dry basis in a conventional liquid supplement. Approximately 40% glycerin was included in the LFS with 12.5% molasses vs typical all molasses based LFS.
- Results
 - Dry matter digestibility not different
 - Ammonia content and yield of microbial Nitrogen not different
 - Total production of Volatile Fatty acids tended to be higher in glycerin supplement with amount of acetic acid lower and butyric acid higher
 - Ph data implies drop post feeding was less with the glycerin based feed. (next slide)
- **Conclusion- replacement of molasses with glycerin should not impact digestive performance of liquid supplements.**

Impact of Glycerin (40%) on rumen culture ph

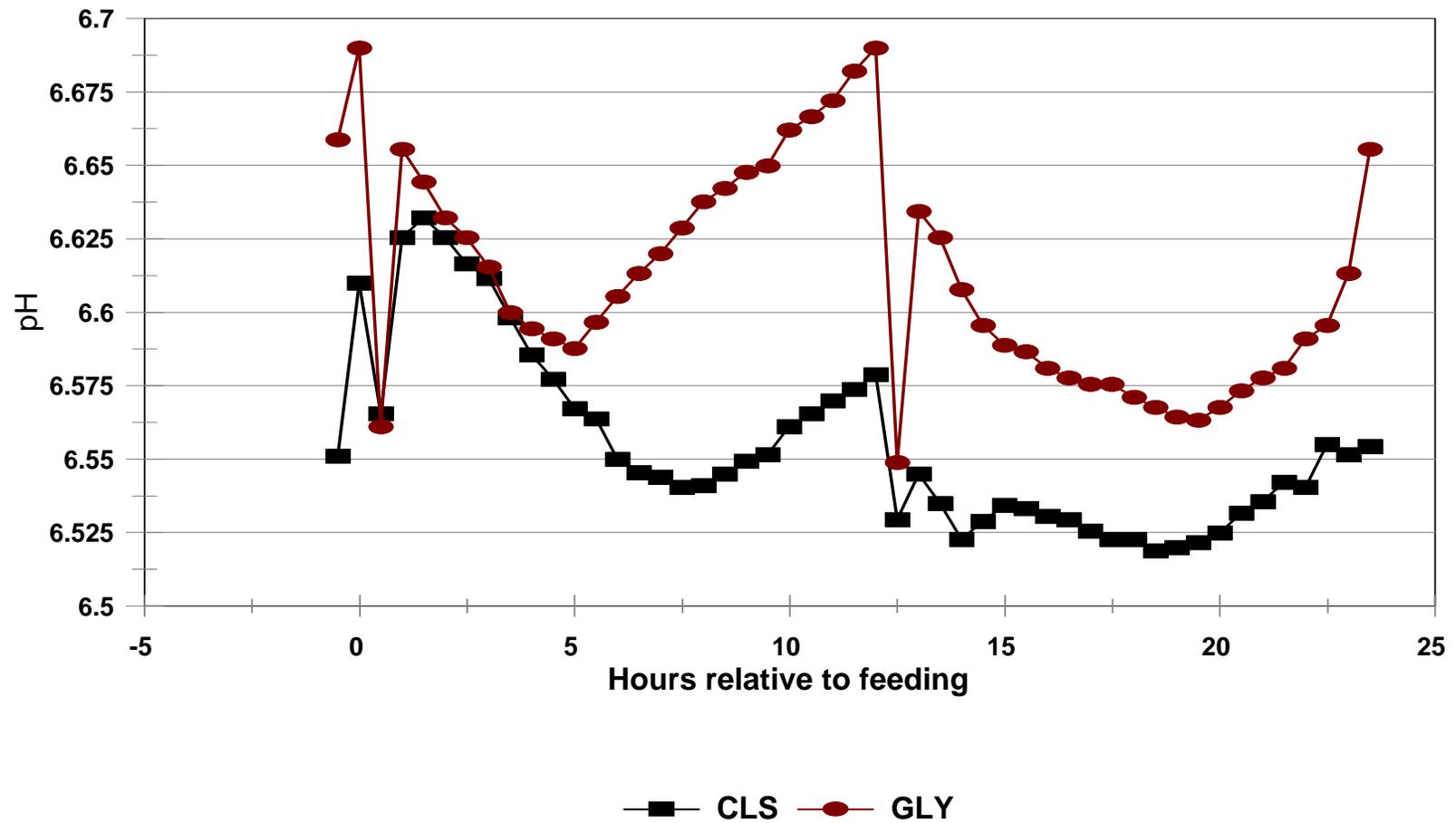


TABLE 5. Effect of Liquid Supplement on Volatile Fatty Acid Production and pH

Component	Treatments		P =
	CLS	GLY	
mM/day :			
Total VFA	181	178	0.73
Acetic	121	107	0.06
Propionic	41	42	0.22
Iso-butyric	0.46	0.55	0.06
Butyric	16	27	0.0008
Isovaleric	0	0	.
Valeric	2.4	2.4	0.93
Molar %			
Acetic	67.1	59.9	0.0011
Propionic	22.5	23.4	0.13
Iso-butyric	0.26	0.31	0.02
Butyric	8.8	15.0	0.0009
Isovaleric	0.0	0.0	.
Valeric	1.3	1.4	0.79

CONSUMPTION STUDIES

Effect of adding USP glycerin to equine feed to determine if feed refusal is an issue:

Added 1% glycerin to normal ration. Fed eight horses - 1 colt, 4 Thoroughbred mares, 1 Arab mare, 1 Arab gelding, 1 Thoroughbred gelding. Ages range from 6 months to 29 yrs.

Everything was cleaned up in normal time. Each horse took the first bite, chewed it up, and went back for the next with no hesitation. Bottom line is 1% glycerin caused no noticeable difference to the horses.

Consumption of high crude glycerin biodiesel origin content liquid supplement by pregnant beef cows confined in a dry lot: Two groups of beef cows were separated into pens in a dry lot. Both groups were offered a liquid supplement that contained 42.43% glycerin (formula shown below) and a diet consisting of cotton seed burrs and a 30% burr ration. One group was fed with a typical lick wheel feeder while the other group was fed using an open trough feeder. The trial period for the consumption study was 18 days. Diet and performance data are summarized below. As expected, the cattle consuming the liquid supplement from the open trough consumed more than the cattle exposed to the lick wheel feeder (10.6 vs. 7.0 lbs/hd/day). From this information it is ascertainable that feed refusal due to glycerin is not an issue. Further more the cows performed well on the product averaging 1.7 lbs/hd/day gain over the 18 day period.

Kenneth Eng, Jr. Ph. D.
Consultant
Eng Inc.

10/25/06

Observation on -C- Cattle at Nolan County Feeders

	Group 1	Group 2
	163 Cows	113 Cows
	18 days	18 days
Burrs	17.0#/ hd/dy	14.1#/hd/dy
30% Burrs Rations	4.2	3.5
Liquid (Liquid Wheel)*	7.0	-0-
Liquid (open)*	-0-	10.6
Total lbs/hd/day	28.2	28.2
Dry Matter/hd/day	24.3	23.2
# Gain/dy	1.7	1.7

Cows Description Medium frame size, solid mouth, Brangus type cows
 Central Texas sale barn origin, Average wt. 1075lbs

Westway Experiment Blend

WESTWAY FEED PRODUCTS
 Solution Report
 Plant : 505 - Hereford
 Pricing Plant : 505 - Hereford

Page : 1

Description	Species	Date	Stored	Current
Code	Code	Code	Code	Code
0015	NOLAN-C COW PREMIX		30	
Amount	Name	Per		
848.5330	014-13 GLYCERIN	* 42.43		
606.5698	001-42 CORN FERM. SOLUBLES	* 30.33		
168.2222	010-35 LIMESTONE	* 8.41		
125.4817	001-09 UREA LIQ 55%	* 6.47		
100.0000	010-19 B-BUFFER	* 5.00		
60.0000	001-98 LIQUID CLAY	* 3.00		
55.8086	001-16 BULK SALT	* 2.79		
23.7957	001-82 18-34-0	* 1.19		
5.0000	003-28 AQUA AMMONIA	* 0.25		
2.0000	012-60 T.M.(GPER)	* 0.10		
0.2500	011-06 CARMEL FLAVOR	* 0.01		
0.1102	002-65 VITAMIN A 400 KI/UG	* 0.01		
0.1102	002-67 VITAMIN E 400 IUG	* 0.01		
0.0965	004-34 SELENIUM 1%	* 0.01		
0.0220	002-46 VITAMIN D 200	* 0.01		
2,000.0000				

Code	Name	Cost	Max.	Low
001-05	CANE MOLASSES-79.5	* 7.2500		4.2656
003-75	MILK DISTILLERS	* 0.9500		0.4040
013-64	SYRUP/MCDS	* 5.4500		3.8035

Nutrient Name	Actual	Dry Matter	Units	Min.	Max.
1 Weight	1.0000	1.0000	Lbs	1.0000	1.0000
2 Dry Matter %	73.0000	100.0000	%	73.0000	100.0000
3 Crude Protein %	16.0000	21.9178	%	16.0000	
4 NPN %	11.3749	15.5821	%		
5 Crude Fat %	0.1228	0.1683	%		
8 Total Sugar Invert	1.3648	1.8696	%		
9 Moisture %	27.0000	36.9863	%		
10 Calcium %	3.0010	4.1110	%	3.0000	3.0010
11 Phosphorus %	0.5000	0.6849	%	0.5000	
12 Salt %	3.0010	4.1110	%	3.0000	3.0010
13 Sodium %	1.1963	1.6387	%		
14 Chloride %	1.7357	2.3777	%		
15 Magnesium %	0.1533	0.2101	%		
16 Potassium %	1.5603	2.1374	%		
17 Sulfur %	0.2673	0.3662	%		
18 Cobalt ppm	1.0152	1.3996	ppm		
19 Copper ppm	25.9499	35.4930	ppm		
20 Iron ppm	236.1303	323.4661	ppm		
21 Iodine ppm	5.0000	6.8493	ppm		
22 Manganese ppm	86.8657	117.8992	ppm		
23 Selenium ppm	1.0000	1.3699	ppm	1.0000	
24 Zinc ppm	118.1971	161.9128	ppm		
26 Vitamin A KIU/lb	10.0000	13.6986	KIU/lb	10.0000	
27 Vitamin D KIU/lb	1.0000	1.3699	KIU/lb	1.0000	
28 Vitamin E IU/lb	10.0000	13.6986	IU/lb	10.0000	
72 TDN %	52.7841	72.1974	%		
75 NE (Lact) Mcal/cwt	55.4750	75.9932	Mcal/cwt		
76 NE (Main) Mcal/c	59.6403	81.6990	Mcal/cwt		
77 NE (Gain) Mcal/cwt	41.5015	56.8514	Mcal/cwt		

User : WALTD
 Set : ** Multiblend **

Tuesday, September 12, 2006
 11:36:44 AM

Methanol Considerations

- Human metabolism includes conversion to formaldehyde and then formic acid-formic acid responsible for toxic effects- in some species excretion via respiration and urine is documented
- CFR 573.460 permits use of formaldehyde in feeds wherein approximately 25 % of animals diet is comprised of a protein meal treated with up to 1% formaldehyde. This would equate to approximately 0.25% dietary formaldehyde. Molecular weight of formaldehyde is 30.03 and molecular weight of methanol is 32. $.25 * (32.04 / 30.03) = .2667$ % Substituting methanol for formaldehyde and assuming 20% glycerin in diet would equate to $.2667 / 20$ or 1.333 % methanol in glycerin source
- CFR 573.480 describes use of formic acid in hay crop silages as a preservative not to exceed 2.25% on dry weight basis- assuming 50% silage on a dry basis in diet would provide 1.125% formic acid. Formic acid m.wt. = 46.02. Methanol m.wt. = 32.04. Adjusting for molecular wt. $1.125 * (32 / 46) = .783\%$. $.783 / 20 =$ would equate to 3.915% methanol in a glycerin source fed at 20% of diet to ruminant animals.
- CFR 573.640 describes the use of “methyl-esters” of higher fatty acids for use in animal feeds. Methyl-esters are considered non-toxic with LD 50 > 17.4 g/kg in rats. Digestion includes the release of methanol from the fatty acid. As a portion of molecular weight assuming C-16/0 as an average- methanol yield equals approximately 11.83% of inclusion of the methyl-esters. If diets contained 5 % methyl esters $5 * 0.1183 =$ methanol contribution (.5915%). Again using a 20% inclusion of glycerin source $.5915 / 20 = 2.957\%$ methanol concentration in glycerin would be equivalent.
- Numerous literature references are available relative to feeding formic acid or its' calcium/potassium salts at levels near 1% on a formic acid basis. Methanol concentrations in order to reach this level and again adjusted for relative molecular weights would equal $1 * (32 / 46) = 0.695$ %. With glycerin content of 20% in diets $.695 / 20 = 3.478\%$ methanol would need to be present to provide these levels.

Methanol Considerations cont.

- EPA, 1994 cites a no observed adverse effect level (NOAEL) of 500mg/kg/day for rats fed 90 days.
 - Assuming a dietary dry matter intake of 2.3 % of body weight (23 grams/kg) a value of $0.5\text{g}/23\text{g} = 2.17\%$ of total diet. If glycerin was source at 20% of diet $2.17/20 = 10.85\%$ methanol concentration tolerable in glycerin.
 - NOAEL level can be extrapolated in number of ways. A factor for interspecies differences could be applied and a factor for sensitive sub-populations could be applied. Assuming a safety factor of 3 for possible species differences and a factor of 10 for possible sensitive sub-populations an acceptable daily intake (ADI) would be calculated as $500/(3 \times 10) = 17$ mg/kg/d

ADI(mg/kg/d) =		17		%Glycerin in Diet =		10	
Species	Age (wks)	bwt (kg)	FI (g/kg bwt)	ppm MtOH (max)			
Chickens, broilers	2	0.3	160	1063			
Chickens, broilers	7	2.1	62	2742			
Chickens layers	20	1.3	46	3696			
Chickens layers	40	1.9	47	3617			
Swine young		4	62	2742			
Swine mature		100	31	5484			
Cattle growing		135	27	6296			
Cattle beef mature		500	20	8500			
Cattle, dairy lactating		600	32	5313			
Horses (concentrate)		500	12	14167			

S.O.P. Development Documentation

WESTWAY FEED PRODUCTS, INC.

**Glycerin Resulting From Bio-diesel Production
In Liquid Feed Products
Summary of Evaluation and Use**

Prepared By: Paul Mostyn, Joe Harris, Dave Caldwell, and Tom Geary

Date: October 30, 2006

Revision Date: 11/1/06

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Proposed Quality / Usage Parameters (Glycerin-Feed Grade)

Chemical Properties:

Moisture	5-25%
Glycerol	> = 95% of the organic matter
Phosphorus	0.3% max in dry matter
Potassium	3% max in dry matter
Sodium	3.9% max in dry matter
Chloride	Guaranteed by supplier
Sulfur	Guaranteed by supplier
Methyl Esters	1% max in dry matter
Fat	1% max in dry matter
Methanol (Method AOAC 973.23 GCFID 16 th ed. 1995)	0.75% max in dry matter
Total Ash	12% max in dry matter
Lead	30 ppm max in dry matter
Cadmium	0.5 ppm max in dry matter
Nickel	50 ppm max in dry matter
Mercury	2.0 ppm max in dry matter
Selenium	2.0 ppm in the dry matter
Arsenic	50 ppm max in dry matter

Proposed Quality/Usage Parameters (cont.)

Glycerin-Feed Grade

Physical Properties:

- Mild pleasant aroma
- Near neutral ph- 5 to 7.0

Usage Guidelines: via Labeling

- Not to be used in combination in diets containing formic acid, formaldehyde and methyl-esters.
- Guarantees for moisture, ash, sodium, potassium, sulfur and chloride maximums
- Limited inclusion rate in feeds i.e. poultry diets (5-10%), equines and swine (10%) and ruminants (20%)

Summary

- Glycerin from bio-diesel production is:
- An energy dense, palatable material for use in feed
- Of high purity-particularly when evaluated on an organic matter basis
- Low in concentrations of heavy metals
- Low in methanol concentration compared when compared levels of either metabolites approved for feed use or published toxicity values
- Can represent a value to animal feeding programs