

# AAFCO AV Limits in Consideration of Current AAFCO PT Results

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# Background

- Analytical Variations (AV): Measure of acceptable variation from the guarantee based on laboratory variance and the inherent variability of a particular analyte
- No publications have been found describing how analytical variations were first calculated
- The original data has also not been located

Table 3. Feed Ingredient Analytical Variations

Ingredient	Method*	AV%	Range
PROXIMATE ANALYSIS			
Moisture	934.01 930.15 935.29	12	3 - 40%
Protein	954.01 976.05 976.06 984.13	(20X + 2)	10 - 85%
Lysine	975.44	20	0.5-4%
Fat	920.39 954.02 932.02	10	3 - 20%
Fiber	962.09	(30X + 6)	2 - 30%
Ash	942.05	(45X + 3)	2 - 88%
Pepsin Digest	971.09	13	
Total Sugar as Invert	925.05	12	24 - 37%
NPN Protein	941.04 967.07	(80X + 3)	7 - 60%
MINERALS			
Calcium	927.02	(14X + 6)	.5 - 25%
"	968.08	10	10 - 25%
"		12	< 10%
Phosphorus	964.06 965.17	(3X + 8)	.5 - 20%
Salt	969.10	(7X + 5)	.5 - 14%
"	943.01	(15X + 9)	.5 - 14%
Fluorine	975.08	40	ppm
Cobalt	968.08	25	.01 - .16%
Iodine	934.02 935.14 925.56	40	ppm
Copper	968.08	25	.03 - 1%
"		30	< .03%
Magnesium	968.08	20	.01 - 15%
Iron	968.08	25	.01 - 5%
Manganese	968.08	30	.01 - 17%
Potassium	975.03 925.01	15	.04 - 8%
Zinc	968.08	20	.002 - 6%
Selenium	969.06	25	ppm
Sodium	n.a.	20	.2 - 4%
	KCP	15	.2 - 4%
VITAMINS			
Vitamin A	974.29	30	1200 - 218,000 IU/lb
Vitamin B <sub>12</sub>	952.20	45	
Riboflavin	970.65 940.33	30	1 - 1500 mg/lb
Niacin	961.14 944.13	25	3 - 500 mg/lb
Pantothenic Acid	945.74	25	4 - 190 mg/lb

\* Method References are from 17th Edition, AOAC Official Methods of Analysis.

# Methods

- Data from AAFCO check sample datasets
  - From 2014-2017
- 53 total datasets with each dataset containing many different analytes
- Individual observations that had a z-score above 3 or were flagged for another error were deleted from the dataset
- Mean, standard deviation, and 2 x Coefficient of variation were calculated for each analyte within each dataset
- 2 x Coefficient of variation was compared to the AV levels in the AAFCO official publication
- Some of the concentrations fell outside the guarantee range established in the official publication

AAFCO Dataset 1		
Lab	Analyte	Concentration
1	Protein	32
2	Protein	34
3	Protein	25
1	Calcium	2
2	Calcium	2.4
1	Fiber	4
2	Fiber	4.1

AAFCO Dataset 2		
Lab	Analyte	Concentration
1	Protein	14
2	Protein	14
3	Protein	15
1	Calcium	11
2	Calcium	10
1	Fiber	16
2	Fiber	20

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AAFCO Dataset i		
Lab	Analyte	Concentration
1	Protein	21
2	Protein	22
3	Protein	23
1	Calcium	5
2	Calcium	8
1	Fiber	12
2	Fiber	17

Results Dataset				
Data	Analyte	Mean	SD	2*CV
Dataset 1	Protein	30.33333	4.725816	0.311592
Dataset 1	Calcium	2.2	0.282843	0.25713
Dataset 1	Fiber	4.05	0.070711	0.034919
Dataset 2	Protein	14.33333	0.57735	0.080561
Dataset 2	Calcium	10.5	0.707107	0.134687
Dataset 2	Fiber	18	2.828427	0.31427
Dataset i	Protein	22	1	0.090909
Dataset i	Calcium	6.5	2.12132	0.652714
Dataset i	Fiber	14.5	3.535534	0.48766

Protein Dataset			
Data	Mean	SD	2*CV
Dataset 1	30.33333	4.725816	0.311592
Dataset 2	14.33333	0.57735	0.080561
Dataset i	22	1	0.090909

Calcium Dataset			
Data	Mean	SD	2*CV
Dataset 1	2.2	0.282843	0.25713
Dataset 2	10.5	0.707107	0.134687
Dataset i	6.5	2.12132	0.652714

Fiber Dataset			
Data	Mean	SD	2*CV
Dataset 1	4.05	0.070711	0.034919
Dataset 2	18	2.828427	0.31427
Dataset i	14.5	3.535534	0.48766

Analyte	Meet Current AV	Out of Concentration Range	Higher than Current AV
Ash	45/52 = 86.5%	2/52 = 3.8%	5/52 = 9.6%
Fat	25/104 = 24.0%	18/104 = 17.3%	61/104 = 58.7%
Fiber	3/152 = 2.0%	28/152 = 18.4%	121/152 = 79.6%
L-Lysine	46/51 = 90.2%	4/51 = 7.8%	1/51 = 2.0%
Moisture	10/52 = 19.2%	1/52 = 1.9%	41/52 = 78.8%
NPN protein	0/5 = 0%	2/5 = 40%	3/5 = 60%
Protein	18/52 = 34.6%	6/52 = 11.5%	28/52 = 53.8%
Total sugar	1/47 = 2.1%	43/47 = 91.5%	3/47 = 6.4%
Calcium	37/52 = 71.2%	5/52 = 9.6%	10/52 = 19.2%
Cobalt	4/50 = 8.0%	0/50 = 0%	46/50 = 92.0%
Copper	34/53 = 64.2%	0/53 = 0%	19/53 = 35.8%
Fluorine	1/2 = 50.0%	0/2 = 0%	1/2 = 50.0%
Iodine	1/5 = 20%	0/5 = 0%	4/5 = 80.0%
Iron	40/52 = 76.9%	0/52 = 0%	12/52 = 23.1%
Magnesium	52/52 = 100%	0/52 = 0%	0/52 = 0%
Manganese	50/52 = 96.2%	0/52 = 0%	2/52 = 3.8%
Phosphorus	40/52 = 76.9%	6/52 = 11.5%	6/52 = 11.5%
Potassium	47/52 = 90.4%	0/52 = 0%	5/52 = 9.6%
Salt	36/52 = 69.2%	16/52 = 30.8%	0/52 = 0%
Selenium	8/53 = 15.1%	0/53 = 0%	45/53 = 84.9%
Sodium	36/52 = 69.2%	16/52 = 30.8%	0/52 = 0%
Zinc	45/52 = 86.5%	0/52 = 0%	7/52 = 13.5%
Niacin	4/12 = 33.3%	0/12 = 0%	8/12 = 66.7%
Pantothenic acid	2/12 = 16.7%	0/12 = 0%	10/12 = 83.3%
Riboflavin	14/39 = 35.9%	0/39 = 0%	25/39 = 64.1%
Vitamin A	1/43 = 2.3%	0/43 = 0%	42/43 = 97.7%
Vitamin B <sub>12</sub>	1/6 = 16.7%	0/6 = 0%	5/6 = 83.3%

# Results

- Fat, fiber, moisture, protein, selenium, riboflavin, and vitamin A were the major analytes who had a large percentage of higher AV.
- A few of the analytes are harder to distinguish due to their small sample size.

# Further Actions

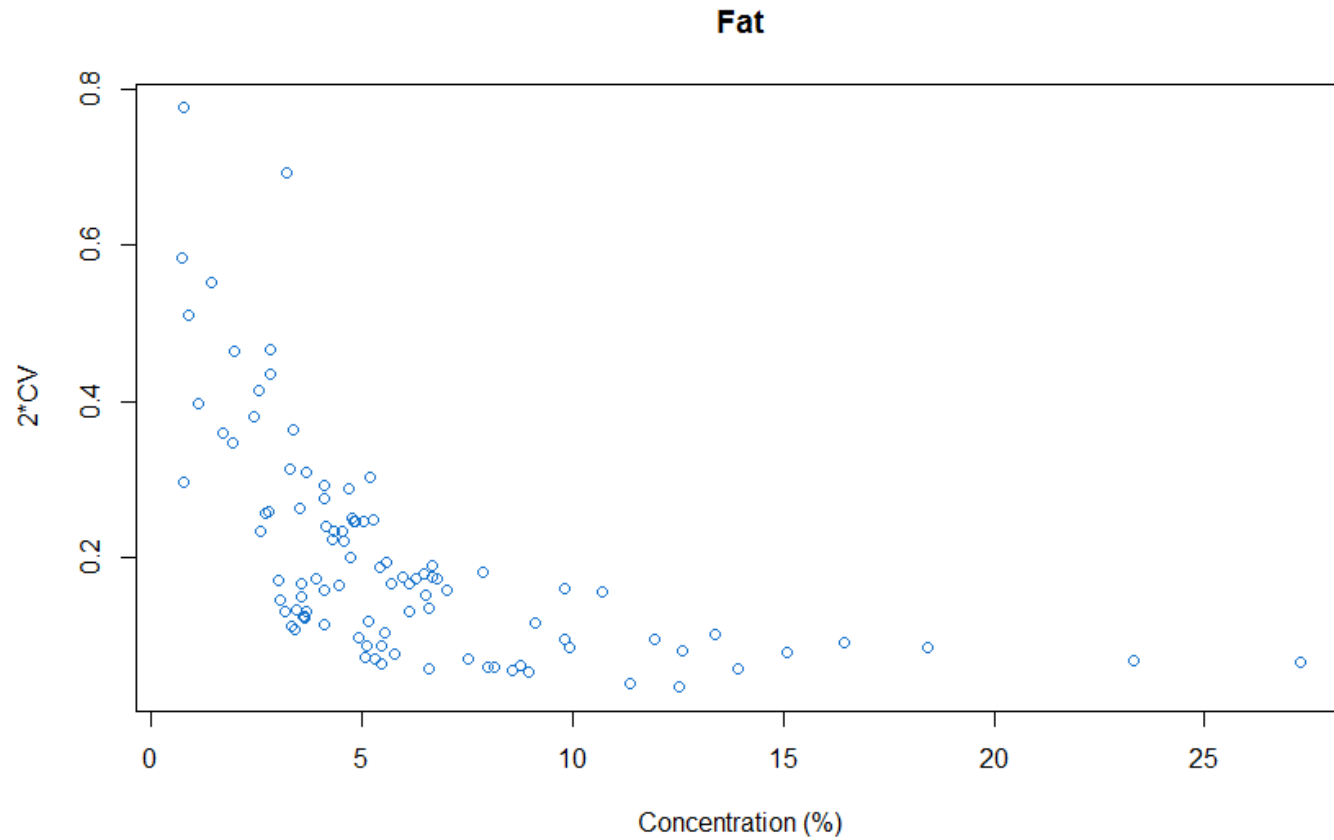
- Created possible changes to the AV using tolerance intervals
  - Tolerance intervals are similar to confidence intervals, but look at the distribution instead of a single parameter like the mean.
  - The tolerance interval used for this was a 95%, 90% interval
- Some of the analytes have AV that are dependent on concentration.
  - Spearman's correlation coefficient was used to determine which ones are dependent.



# Spearman's Correlation Coefficients

Analyte	Spearman's Correlation
Ash	-.331
Cobalt	-.586
Moisture	-.658
Phosphorus	-.189
Calcium	-.347
Copper	-.842
Fat	-.726
Fiber	-.598
Iron	.272
Lysine	-.301
Magnesium	-.168
Manganese	-.248
Potassium	-.434
Protein	-.745
Selenium	-.359
Zinc	-.376

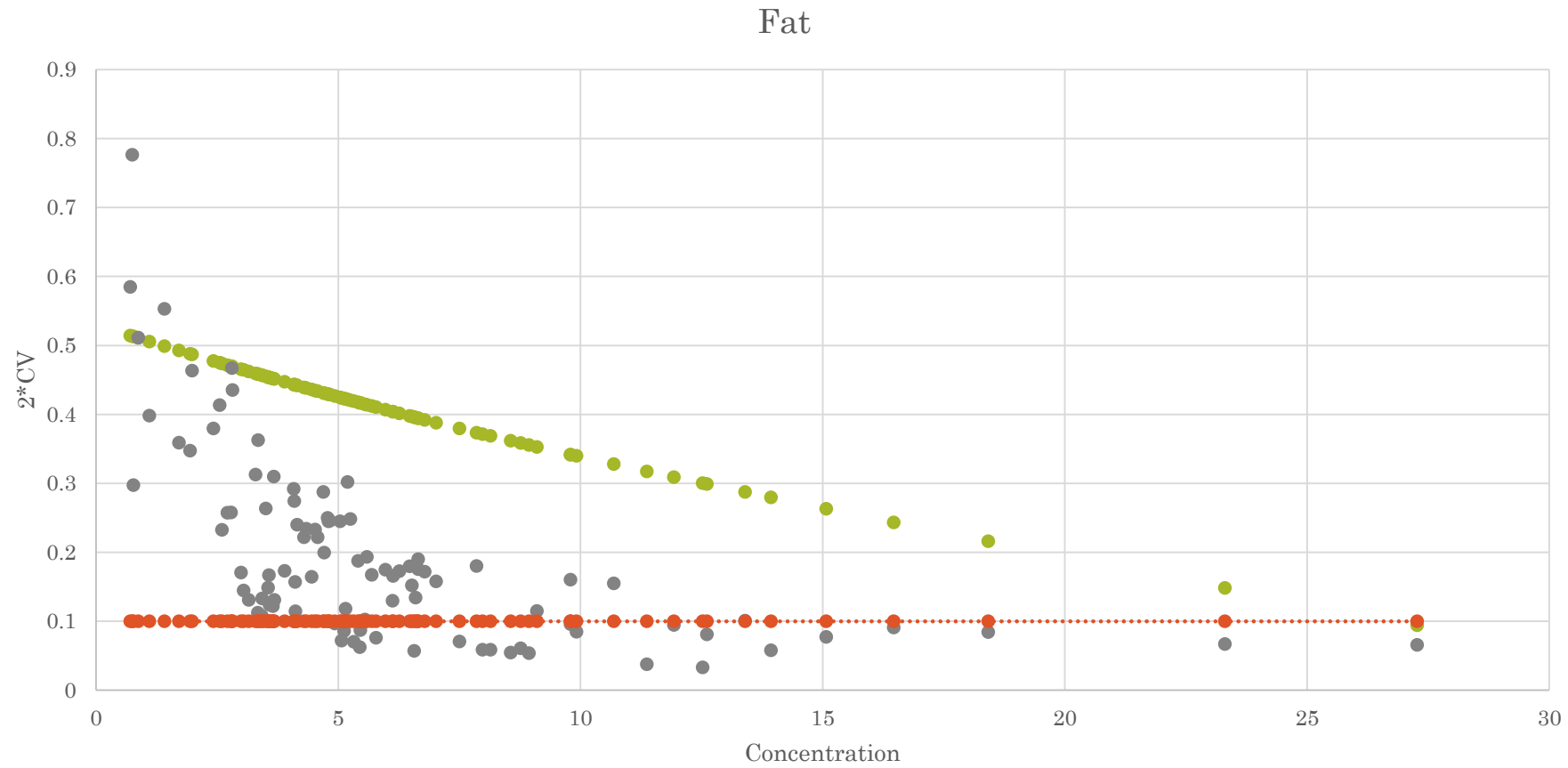
# Example - Fat



# Example - Fat

- Check Spearman's correlation coefficient
  - $-.726$
- Concentration dependent so a linear regression model was fit
- Q-Q plot was used to check for the normality assumption
- The 95% tolerance interval was found using the regression line
  - $AV = 55.6e^{-.054*x}$

# Example - Fat



# Computed AV

Analyte	New AV (%)	Current AV (%)
Ash	$9.9e^{-.005*x}$	$(45/x + 3)$
Moisture	$56.9e^{-.079*x}$	12
Cobalt	$86.1e^{-.008*x}$	25
Phosphorus	13	$(3/x + 8)$
Calcium	$13.6e^{-.0006*x}$	$(14/x + 6)$
Fat	$55.6e^{-.054*x}$	10
Fiber	$50.9e^{-.02*x}$	$(30/x + 6)$
Iron	33	25
Lysine	16	20
Magnesium	15	20
Manganese	21	30
Potassium	$18.5e^{-.08*x}$	15
Protein	$4.6e^{-.006*x}$	$(20/x + 2)$
Selenium	$61.9e^{-.002*x}$	25
Zinc	$26.5e^{-.00002*x}$	20