STANDARDIZATION BY INTERNAL STANDARD METHOD

OBJECTIVE

To illustrate a technique for quench correction.

THEORY

Since it is sometimes desirable to be quantitative when performing a nuclear assay, a means for determining the relationship between quench and counting efficiency must be determined. We know, for example, that as we increase attenuation of the photon output of the sample or quench the sample, the ratio of cpm to dpm gets small. Our objective then is to make an exact determination of this relationship so that dpm can be calculated knowing only cpm and the numerical relationship referred to above. The following technique solves the problem! A known amount of dpm is added to the sample after it has been counted. The sample is then counted again and the incremental increase in cpm is divided by the dpm that was added, thus yielding the decimal efficiency. The original cpm is then divided by decimal efficiency which then computes the dpm of the sample. Equation (1) is a mathematical statement of the process:

$$\frac{(cpm_1 + cpm_2) - cpm_1}{dpm_2} = E \qquad (1)$$

$$\frac{cpm_1}{E} = dpm \qquad (2)$$

E = decimal efficiency

cpm_1 = unknown sample

cpm_2 = increase due to the addition of dpm_2

dpm_1 = true activity of sample

dpm_2 = activity added to sample

MATERIALS

1. 6 scintillation vials - 1" x 2 1/2" (Low K)
2. 90 mls scintillation fluid whose composition is:
   a. 8 gms/l PPO = 2, 5 - diphenyloxazole
   b. 0.4 gms/l dimethyl POPOP = 1, 4 - bis[2-(4-methyl-5-phenyloxazolyl)]-benzene
   c. Toluene to volume
3. Several microliters of activity bearing solution:
   a. ^{3}H labeled toluene

PROCEDURE

1. Prepare six samples with varying quench, and at an activity level as indicated in Table I.

TABLE I

Sample #	Toluene + Fluors	Nitro Methane	^{3}H (dpm added)	Internal Standard (dpm added)
1	12 ml	0.005	30,000	30,000
2	12 ml	0.015	30,000	30,000
3	12 ml	0.025	30,000	30,000
4	12 ml	0.035	30,000	30,000
5	12 ml	0.045	30,000	30,000
6	12 ml	0.055	30,000	30,000

2. Count each sample and record in the space provided in Table II.
3. Add the dpm as indicated in the last column of Table I to each sample, recount each sample and complete Table II.

TABLE II

	A	B	C	D
Samp. #	Sample cpm	Sample + added cpm	B-A/dpm added	A/C = dpm Sample
1				
2				
3				
4				
5				
6				

QUESTIONS

1. List advantages and disadvantages of this technique.*
2. How would you improve this technique and/or calibrate for its shortcomings?

* Consider the effect of variation in mole fraction of fluors, solvent and sample.

OPTIONAL EXPERIMENT

1. Repeat this experiment with a set of unknown samples supplied by the instructor.

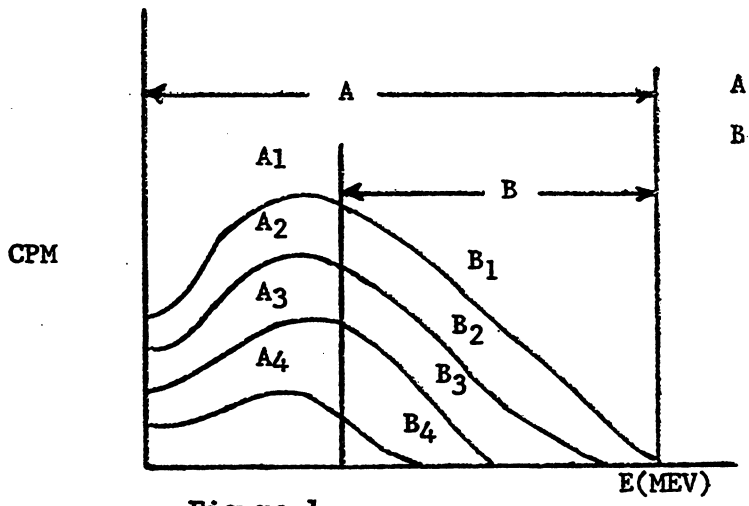
STANDARDIZATION BY CHANNELS-RATIO METHOD

OBJECTIVE

To illustrate a technique for quench correction

THEORY

This method attempts to correct for quenching of the photon output of the sample by following the change in the ratio of sample counts falling in two counting channels and the corresponding change in efficiency.



A = Counting channel

B = Extra channel for use in channels ratio quench correction

Figure 1.

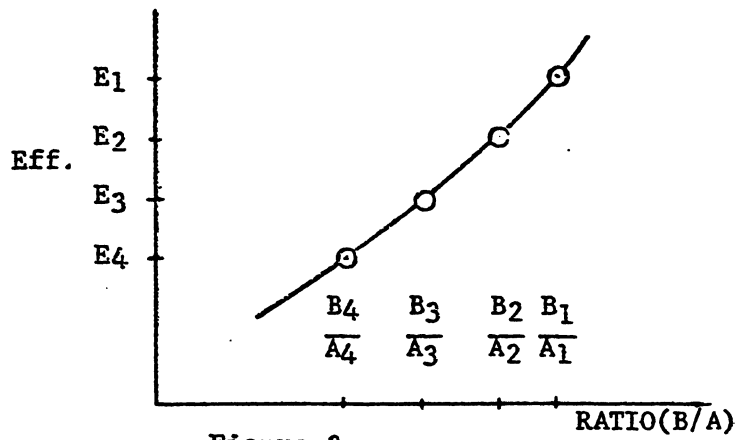


Figure 2.

Figure 1 shows the count distribution in two channels, labeled A and B. Several examples of a quench count distribution are also shown. It is clear that the ratio of count in Channel A to those in Channel B will decrease as quenching increases. By definition, efficiency decreases as quenching increases.

Figure 2 is a graphic combination of these facts. Such a curve is generated from data gleened from a set of standards, where each member of the set has a different amount of quenching agent and a known amount of activity.

When samples of unknown dpm are counted and an attempt to correct for quench is made by this method, the windows used to set up the calibration curve must also be used for the sample count.

MATERIALS

1. 12 scintillation vials - 1" x 2 1/2" (Low K)
2. 180 ml scintillation fluid whose composition is:
   a. 8 gms/l PPO = 2, 5 - diphenyloxazole
   b. 0.4 gms/l dimethyl POPOP = 1, 4 - bis[2-(4-methyl-5-phenyloxazolyl)]-benzene
   c. Toluene to volume
3. 2 ml CH_3NO_2
4. Several microliters of labeled solution
   a. ^3H labeled toluene

PROCEDURE

1. Prepare standards as indicated in Table I.

TABLE I

Std. #	Scintillation Fluid	CH_3NO_3 (ml)	^3H dpm
1	15 ml	0.005	30,000
2	15 ml	0.010	30,000
3	15 ml	0.015	30,000
4	15 ml	0.020	30,000
5	15 ml	0.025	30,000
6	15 ml	0.030	30,000

2. Obtain 5-6 samples from instructor, which have varying amounts of both quencher and activity.

TABLE II

Std. #	A Wide Open Window CPM	A/dpm	B 70% Window CPM	B/A
1				
2				
3				
4				
5				
6				

3. Count the standards in two different windows, see Figure 1, and fill in Table II.

NOTE: For 70% window, maintain the upper discriminator at a constant level and vary only the lower discriminator to obtain 70% discrimination.

4. Plot A/dpm (Efficiency) vs. B/A.

5. Count the unknown samples obtained from the instructor in the same two windows in which the standards were counted, and fill in Table III. The last column, dpm, is computed by using the Efficiency vs. B/A plot from #4.

TABLE III

Samp. #	A Wide Open Window CPM	B 70% Window CPM	B/A	Efficiency	dpm
1					
2					
3					
4					
5					

QUESTIONS

1. List advantages and disadvantages of this correction method.*

* Consider a count at low levels where the counting error is to be maintained at a constant level.