

# <sup>67</sup>Ga Double Spectral Index Plots and Their Applications to Quench Correction of Mixed Quench Samples in Liquid Scintillation Counting

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## INTRODUCTION

To prepare samples suitable for liquid scintillation counting various materials are added to scintillator solutions. These materials and the sample itself will alter the counting efficiency to some degree; this effect is known as quenching. Chemical quenching arises in an energy transfer process prior to creating luminescence, whereas color quenching is a phenomenon occurring after the luminescence production. Color quenching effect diminishes the mean free path of the fluorescence photons. It is emphasized that chemical quenching is really a physical effect rather than a chemical effect.<sup>1</sup> The pulse height distribution of a color quenched sample differs from that of a chemically quenched sample, even if both samples have identical radioactivity and counting efficiencies. Therefore, chemical and color quench correction curves are also different. For strongly quenched samples there is a large difference between the two kinds of quench correction curves.<sup>2</sup> Significant errors can be encountered if a quench correction curve obtained from chemically quenched standards is applied to color quenched samples. Thus, it is advantageous to determine the type of quench in a sample before quench correction is applied.

Let us discuss some mixed quench samples where quenching is produced by the mixed solutions of a given chemical quencher ( $\text{CCl}_4$ ) and a color quenching solution at different ratios. Since the counting efficiency of the sample with known radioactivity (dpm) can be calculated, its quenching type is easily determined according to the location on the chemical and color quench correction curves. This method cannot be used, however, if sample activity is unknown. Therefore, a new parameter, quench type contribution factor (QTCF), was established to denote the degree of contribution from chemical or color

quenching in a mixed quenched sample.<sup>3,4</sup> The mixed quenched sample could be corrected automatically by means of the chemical and color quenched standard samples, and QTCF value, and an on-line computer. Some authors have used the double ratio technique to determine the quench type.<sup>5</sup> This method was first used by Bush in 1968 to detect adsorption or precipitation of radioactive material from liquid scintillation solutions.<sup>6</sup> Earlier, an isolated internal standard method<sup>7</sup> and four curve method<sup>8</sup> were also tried for this purpose.

This paper describes a method called *<sup>67</sup>Ga Double Spectral Index Plots*, which can determine quench type of unknown samples and correct the quenching of mixed quench samples. A novel report about using <sup>67</sup>Ga as an internal standard in liquid scintillation counting was presented by McQuarrie and Noujaim in 1983.<sup>9</sup> Gallium is an element in Group IIIa of the periodic table. The physical half life of the radionuclide <sup>67</sup>Ga is 78 hours with major gamma emissions at 93 (40%), 184 (24%), and 296 (22%) keV. A variety of radiopharmaceuticals have been developed in the past two decades in an attempt to obtain tracers for tumor imaging, among these <sup>67</sup>Ga-citrate has found widespread use<sup>10</sup> thus, <sup>67</sup>Ga can be obtained easily from clinical departments for use in liquid scintillation counting.

## MATERIALS AND COUNTING CONDITIONS

- scintillator solution: aquasol-2, a commercially available xylene based cocktail (New England Nuclear)
- chemical and color quenchers: carbon tetrachloride (CCl<sub>4</sub>), saturated solution of methyl red (reagent grade) in acetone (reagent grade), a specially prepared reagent. All quenchers and their solvents, products of Nakarai Chemicals Ltd. (Japan)
- labeled compounds: <sup>67</sup>Ga-citrate injection (Medi-physics Co. Japan) as <sup>67</sup>Ga source, Thymidine (6-<sup>3</sup>H), > 15 Ci/mmol, aqueous solution (New England Nuclear), n-(1,2(n)-<sup>3</sup>H)hexadecane (TRR-6) and n-(1-<sup>14</sup>C)hexadecane (CFR-6), manufactured by Amersham International, used as standards in preparations of <sup>3</sup>H and <sup>14</sup>C quenched series
- instrument and counting conditions: Packard Tri-Carb (Model No.460CD) liquid scintillation spectrometer; spectral index (SIS and SIE), sample channels ratio (SCR), and spectra, as counting conditions used as listed in Table 1

All counting was done at 16°C and all samples were contained in 20 mL standard glass vials in which 10 mL cocktail was added.

## EXPERIMENTS AND RESULTS

### Quenched <sup>67</sup>Ga Spectra

A series of quenched samples containing the same amount of <sup>67</sup>Ga-citrate (5 μL aqueous solution) and varying amounts of carbon tetrachloride (chemical

**Table 1. The Selection of Counting Conditions**

	Energy Range (keV)	SCR	Spectra (keV)	
			Window Width	Range
<sup>3</sup> H	0-19	counts (4-19 keV) counts (0-19 keV)	1	0-20
<sup>14</sup> C	0-156	counts (10-156 keV)	1	0-20
		counts (0-156 keV)	5	21-160
<sup>67</sup> Ga	0-3,5,7,10,19,156	counts (0-19 keV)	1	0-20
	0-variable upper limit 10-100,15-100	counts (0-156 keV)	5	21-156

quencher) were prepared and tested. Increasing the amounts of the quencher caused a shift of the observed <sup>67</sup>Ga pulse height spectrum in the direction of lower energy (Figure 1). These spectra of <sup>67</sup>Ga are characterized by two peaks, i.e., lower and higher energy peaks corresponding to Auger electrons at 8 keV and conversion electrons from the 90 keV level, respectively. Though the spectral shift of both lower and higher energy peaks is concentration dependent, the counts at the lower energy peak were sequentially reduced by increasing the quencher. In contrast, the counts at the higher peak were sequentially increased. This response is similar to the spectral shift of <sup>3</sup>H and <sup>14</sup>C quenched series (Figure 2). The energies of two peaks of <sup>67</sup>Ga are closely related to the average beta decay energies of <sup>3</sup>H and <sup>14</sup>C, thus, a single or dual labeled sample containing <sup>3</sup>H and/or <sup>14</sup>C would behave similarly to the corresponding peaks of <sup>67</sup>Ga at the same level of sample quench. McQuarrie and Noujaim<sup>9</sup> developed a technique using parameters related to the pulse height and number of events in each peak of <sup>67</sup>Ga. With it they may obtain a mathematical relationship that monitors the degree of sample quench.

**Spectral Index of <sup>67</sup>Ga**

Spectral index of samples (SIS) and spectral index of external standard (SIE) are two of the current quench monitoring techniques. The spectral index is a unique number for the given total spectrum and is related to the radionuclide and its level of quenching. If only a part of the spectrum is analyzed, however, this number will not be unique and will depend on the counting conditions. This last point has not attracted sufficient attention until now. Variations of SIS in an unquenched <sup>67</sup>Ga sample for various pulse height ranges (i.e., energy range) are shown in Figure 3. With a region of interest set between a lower limit of 0 keV and an upper limit of 10 to 30 keV, a plateau is seen on the curve; if the upper range is greater than 60 keV, a second plateau is seen. Both lower and higher energy plateaus are related to two peaks of <sup>67</sup>Ga pulse height spectrum. The <sup>67</sup>Ga SIS values determined in the same region of interest will vary as the quenching increases (see Figure 4).

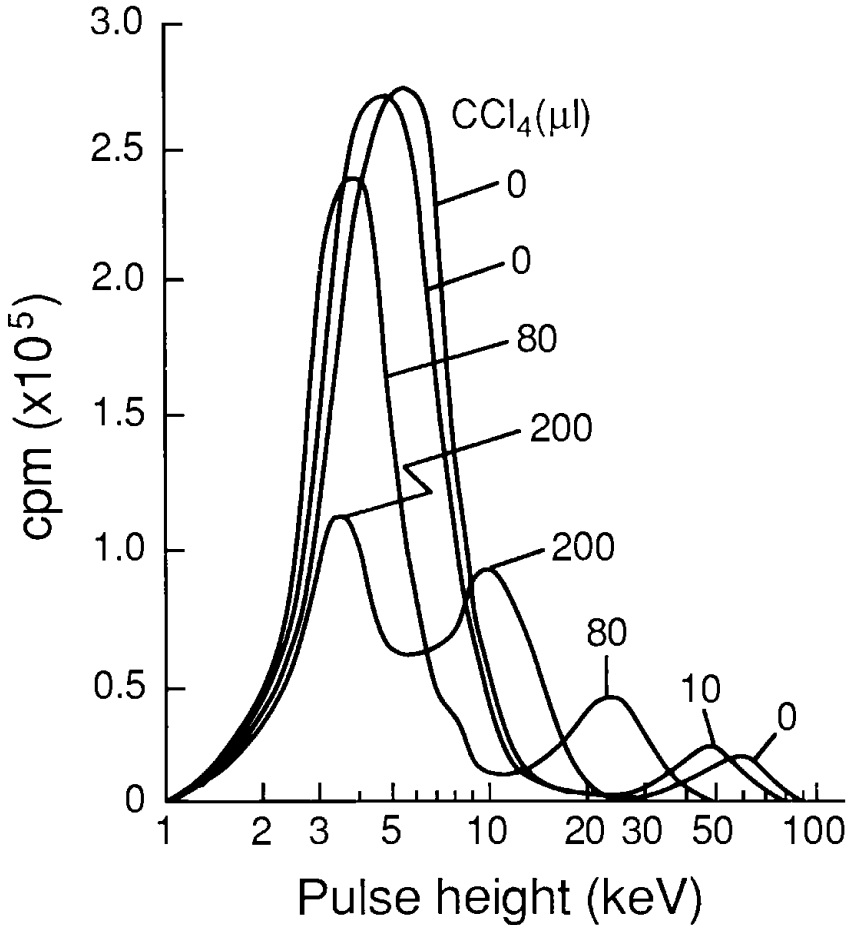
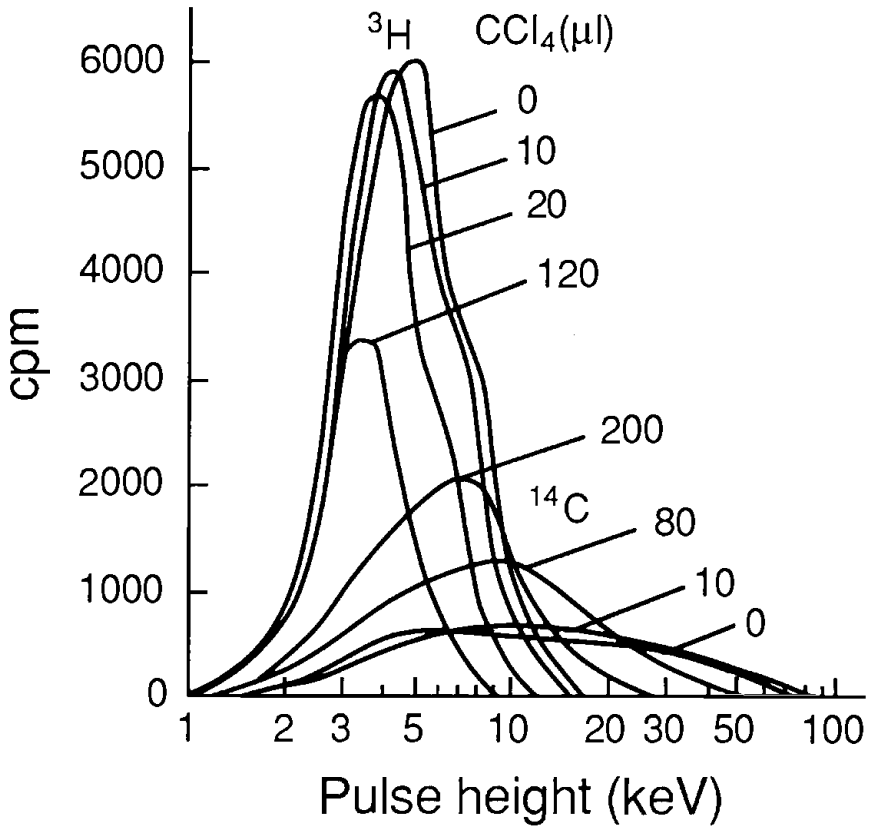


Figure 1. The unquenched and quenched (by carbon tetrachloride) spectra of  $^{67}\text{Ga}$ .

### Double Spectral Index Plots for $^{67}\text{Ga}$

The plot of SIS (0 to 100 keV) vs SIE for chemically quenched  $^{67}\text{Ga}$  is shown in Figure 5. The differences between the shape of chemical vs color quenched spectra of  $^{67}\text{Ga}$  were also obtained (Figure 6); thus, an isolative curve from a chemical quenching curve was obtained for color quenched sample series that contained the same amounts of  $^{67}\text{Ga}$ -citrate (5  $\mu\text{L}$  aqueous solution) and varying amounts of color quenching solution (from 0 to 20  $\mu\text{L}$ ).

A plot of SIS in the lower energy region vs that of SIS in the higher energy region can be obtained when two regions of interest are used to determine  $^{67}\text{Ga}$  SIS values for both chemical and color quenched series, the series must include the lower and higher energy peaks of  $^{67}\text{Ga}$  (Figure 7a). Because the peaks of the color quenched sample were lower and broader in the higher energy side than

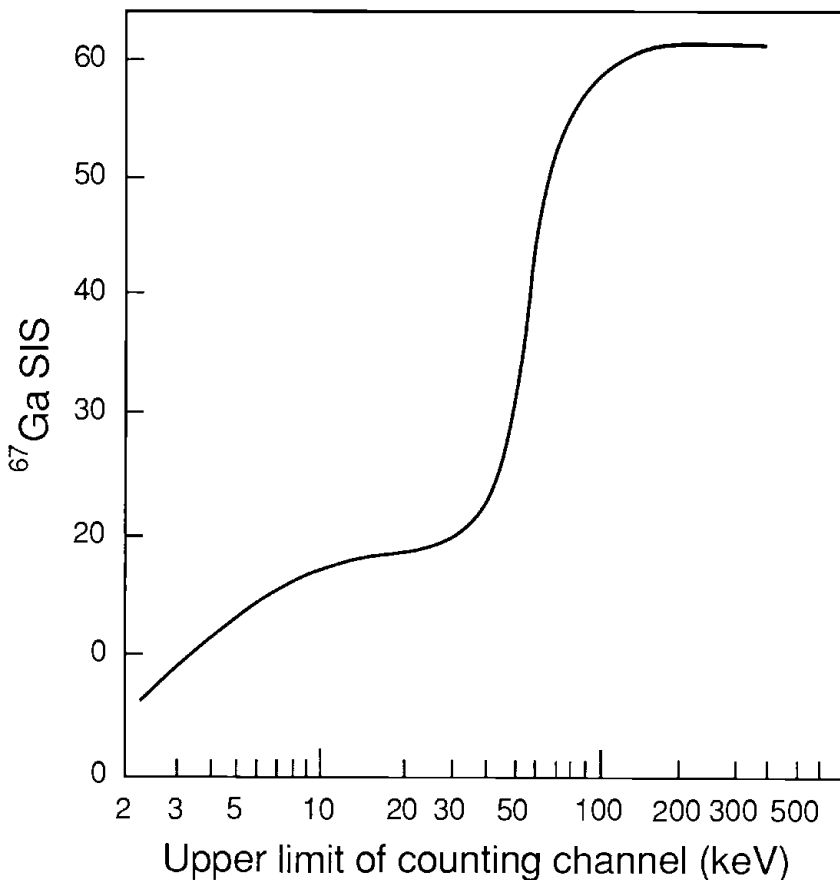


**Figure 2.** The unquenched and quenched (by carbon tetrachloride) beta spectra of  $^3\text{H}$  and  $^{14}\text{C}$ .

the peaks of a chemically quenched sample for  $^{67}\text{Ga}$  (Figure 4), in the same way as in Figure 5, two separate curves were given for chemical and color quenched series (Figure 7a). There is an inflection point on each curve and a crossover point at moderate quenching. When the regions of interest were changed to 7 to 100 keV and 0 to 19 keV, new curves were obtained (Figure 7b) in which there are two inflection points on each curve and a crossover point at a higher level of quench.

### Mixed Quench Zone

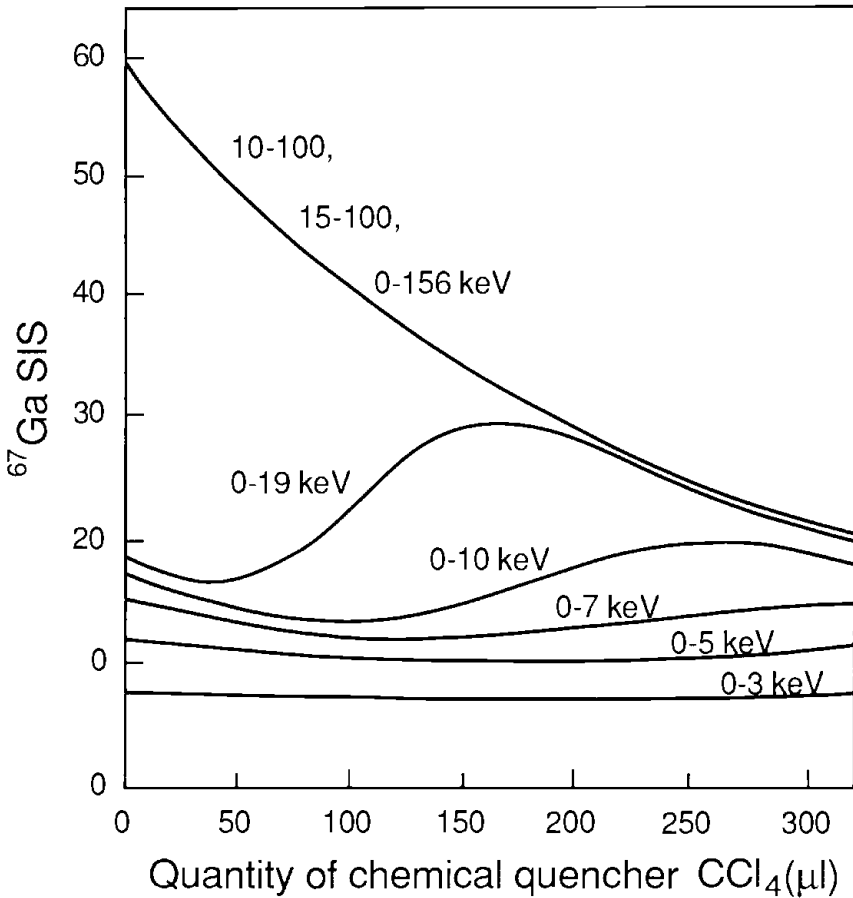
Let us analyze the region between the chemical and color quenched curves in Figures 5, 7a, and 7b. Obviously, these are transitional regions in which samples will simultaneously contain both chemical and color quench; they are referred to as a mixed quench zone. In fact, we prepared samples containing



**Figure 3.** The variation of SIS of unquenched  $^{67}\text{Ga}$  sample in various pulse height ranges.

varying amounts of chemical quenching agent (carbon tetrachloride) and color quenching agent (saturated solution of methyl red in acetone) (Table 2) and measured them at the counting conditions of Figures 5, 7a, and 7b.

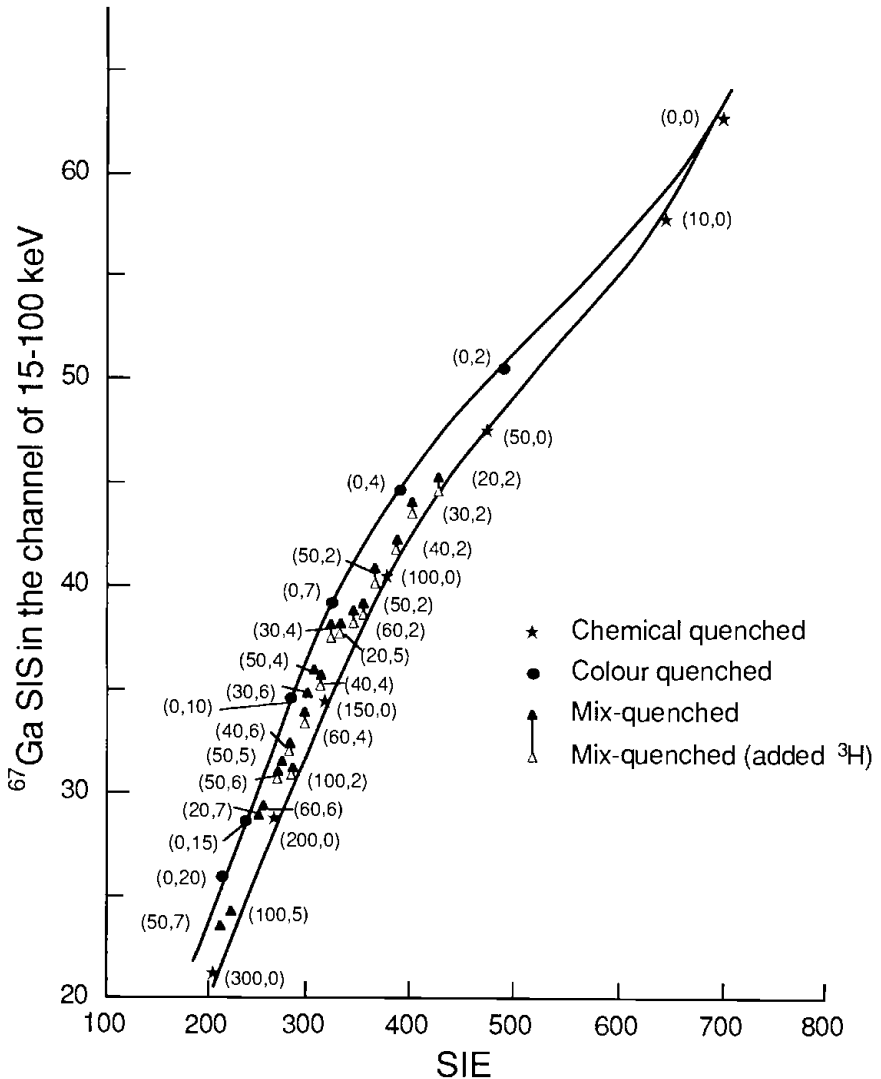
These points of mixed quench samples fall in a zone; the positions in the zone are generally near chemical or color quenching curves, according to their ratio of chemical to color quenching. Similar phenomena have been observed and analyzed in the quench correction curves of sample channels ratio (SCR) and SIS, i.e., the curves of counting efficiency vs SCR or SIS.<sup>3,4</sup> Thus, we can identify the quench type of an unknown sample and calculate the degree of contribution of chemical or color quenching according to its location on the double spectral index plot (Figures 5, 7a, and 7b.) For example, if the quenching agent of a sample is not known,  $^{67}\text{Ga}$  is added to the sample and measured at the above counting condition. A point in double spectral index plots can be obtained from the observed SIS and/or SIE values of the unknown sample.



**Figure 4.** The variations of  $^{67}\text{Ga}$  SIS with increasing quench.

The sample was quenched by chemical quenching when the point is on the chemical quench curve and, in contrast, by color quenching when the point was on the color quench curve. Those samples corresponding to points lying in a mixed quench zone can be considered to contain both chemical and color quenchers simultaneously.

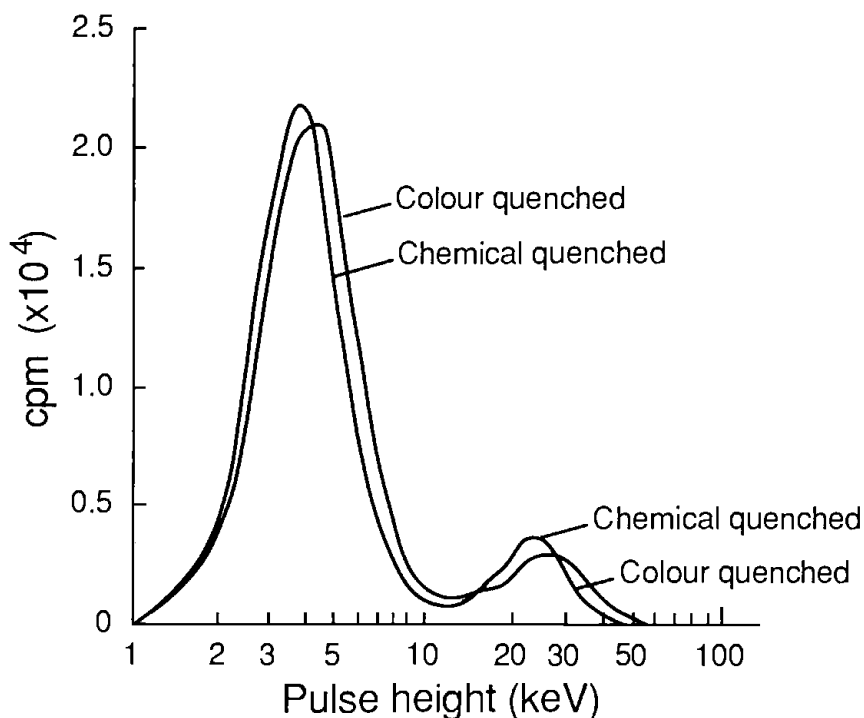
The mixed quench zone was a function of counting conditions in the same way as double spectral index plots. Attention should be paid to the points located near the crossover. Two points [(40,2) and (50,2)] fall outside of the mixed quench zone in Figure 7a, and one point (30,6) is on the color quenched curve in Figure 7b. When counting conditions were changed the mixed quench zone also changed, and these points fell in the new mixed quench zone. Generally speaking, these double spectral index plots are not sensitive to the degree of quench near their crossover point.



**Figure 5.** Double spectral index plots of  $^{67}\text{Ga}$  SIS in the channel of 15 to 100 keV vs SIE. The figures of the first and second groups in the parentheses represent the amounts ( $\mu\text{L}$ ) of chemical quencher ( $\text{CCl}_4$ ) and color quencher (saturated solution of methyl red in acetone).

### Reproducibility of the Point Positions for Mixed Quench Samples

We have given two kinds of double spectral index plots for  $^{67}\text{Ga}$ , i.e., SIS vs SIE, and SIS in a lower energy region of interest vs SIS in a higher energy region of interest. Generally, these two kinds of curves also can be used as a tool to identify the quenching type of an unknown sample. A more important

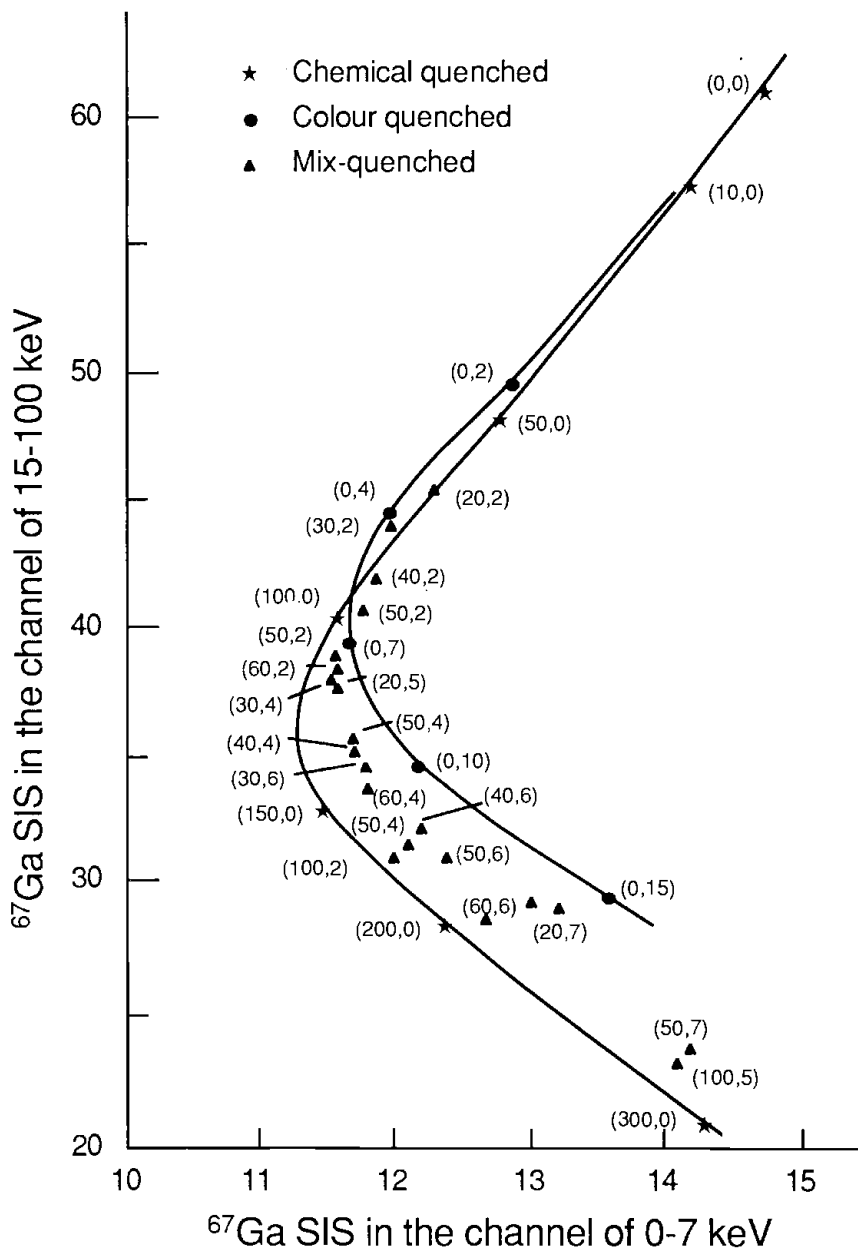


**Figure 6.** The differences of the shapes of  $^{67}\text{Ga}$  pulse height spectra for chemical and color quenching samples containing the same degree of quenching (relative counting efficiency was 68%).

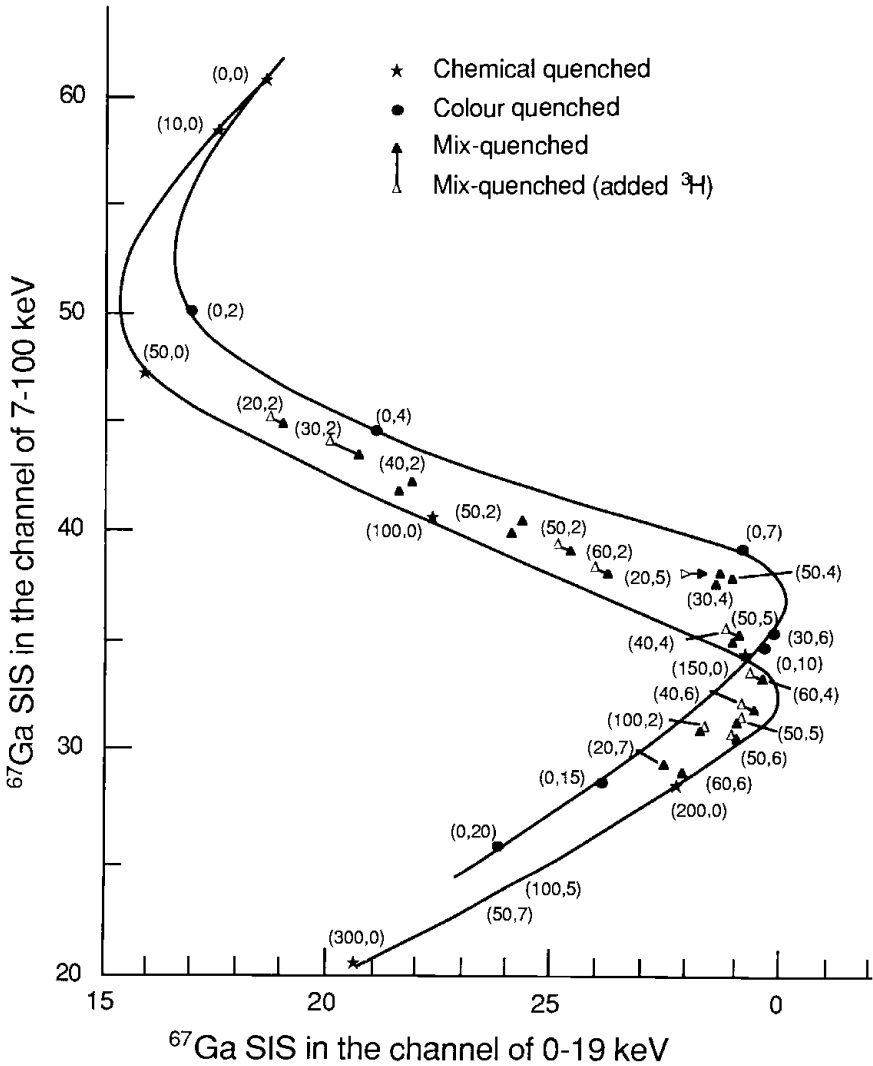
problem for mixed quench samples is the reproducibility of the position in the mixed quench zone, because the zone is smaller than the total region of quench variation.

The main factors affecting the reproducibility of this point are: (1) variation of  $^{67}\text{Ga}$  activity, (2) interference from  $^3\text{H}$  and/or  $^{14}\text{C}$ , and (3) selection and reproducibility of counting conditions. The selection of counting conditions has been discussed under "Spectral Index of  $^{67}\text{Ga}$ ." The reproducibility of counting conditions in this instrument are satisfactory as they are set automatically. To observe the effect of  $^{67}\text{Ga}$  radioactivity on the reproducibility of SIS, a series of quenched samples with three different quantities of  $^{67}\text{Ga}$  (5, 10, and 20  $\mu\text{L}$ ) were prepared and measured repeatedly. The standard deviations of measurements caused by the variation of  $^{67}\text{Ga}$  activity were  $(0.60 \pm 0.20)\%$  (mean  $\pm$  S.D.) for the region of interest of 0 to 7 keV and  $(1.14 \pm 0.66)\%$  for the region of interest 15 to 100 keV which were similar to the standard deviation for 10 measurements of a single sample were similar  $(0.80 \pm 0.26)\%$ . There was no significant difference between either. Changing the quantity of  $^{67}\text{Ga}$  from 5 to 20  $\mu\text{L}$  affected the reproducibility of SIE  $2.1 \pm 0.8\%$ .

If unknown samples containing  $^3\text{H}$  and/or  $^{14}\text{C}$  only make these points shift a



**Figure 7a.**  $^{67}\text{Ga}$  double spectral index plots of SIS in the channels of 15–100 keV to 0–7 keV. The illustration of the figures in the parentheses is shown in Figure 5.



**Figure 7b.**  $^{67}\text{Ga}$  double spectral index plots of SIS in the channels of 7–100 keV to 0–19 keV. The illustration of the figures in the parentheses is shown in Figure 5.

little (Figures 5, 7a, and 7b), their effect on the identification of quenching type and on quench correction can be ignored, because  $^{67}\text{Ga}$  has a sufficiently high counting rate. If  $^3\text{H}$  and/or  $^{14}\text{C}$  in the samples have higher counting rates, usual double ratio techniques<sup>5</sup> and SCR quench correction methods<sup>3,4</sup> can also be used to identify and correct the quench.

Table 2. The Components and Parameters of Mixed Quench Samples

Batch	CCl <sub>4</sub> (μL)	Color Solution <sup>a</sup> (μL)	Equivalent Counting Efficiency (%)		QTCF <sup>b</sup>	F <sup>c</sup>
			<sup>3</sup> H	<sup>14</sup> C		
1	20	2	63.5	95.0	0.615	0.407
		5	34.2	85.6	0.381	0.219
		7	22.7	77.7	0.313	0.185
	50	2	47.9	92.6	0.706	0.607
		5	25.8	83.5	0.353	0.385
		7	17.1	75.8	0.278	0.337
	100	2	28.1	85.9	0.892	0.724
		5	15.2	77.4	0.695	0.516
		7	10.1	70.3	0.497	0.464
2	30	2	57.8	94.6	0.308	0.500
		4	38.4	88.7	0.273	0.327
		6	25.8	80.8	0.188	0.267
	40	2	52.8	93.6	0.759	0.564
		4	34.8	87.8	0.523	0.360
		6	23.5	80.0	0.417	0.320
	50	2	47.9	92.6	0.531	0.607
		4	31.8	86.9	0.445	0.428
		6	21.4	79.1	0.333	0.359
60	2	43.7	91.2	0.625	0.639	
	4	29.0	86.0	0.488	0.462	
	6	19.6	78.3	0.387	0.392	
F = 0.1296 ± 0.6280 QTCF			r = 0.7967			

<sup>a</sup>Saturated solution of methyl red in acetone.

<sup>b</sup>Experimental QTCF of chemical quench from Figure 3.

<sup>c</sup>Calculated contribution factor (F) of chemical quench from <sup>3</sup>H.

## QUANTITATIVE QUENCHING CORRECTION OF MIXED QUENCHED SAMPLES

The color quencher used in this study was a solution of methyl red in acetone (Table 2), but the acetone itself also is a chemical quenching agent. In order to decrease the effect of chemical quenching of acetone, we prepared a saturated solution of methyl red in acetone. The maximum quantity of the color solution used in the work was 20 μL, which decreased the relative counting efficiency of <sup>3</sup>H to 3.5%, and that of <sup>14</sup>C to 32% (Figure 8), relative to the unquenched sample. Under this condition the relative quenching contribution factor (F<sub>c</sub>) of chemical quenching of acetone was only about 0.04 for <sup>3</sup>H. The formula for calculating F<sub>c</sub> factor is as follows:

$$F_c = \frac{1 - E_{rc}}{(1 - E_{rc}) + (1 - E_{rc1})} \quad (1)$$

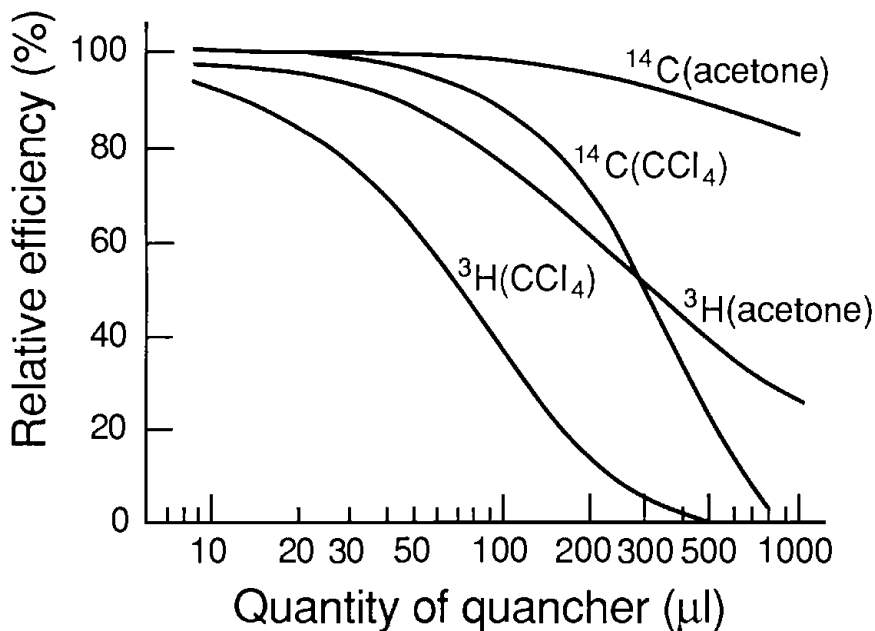


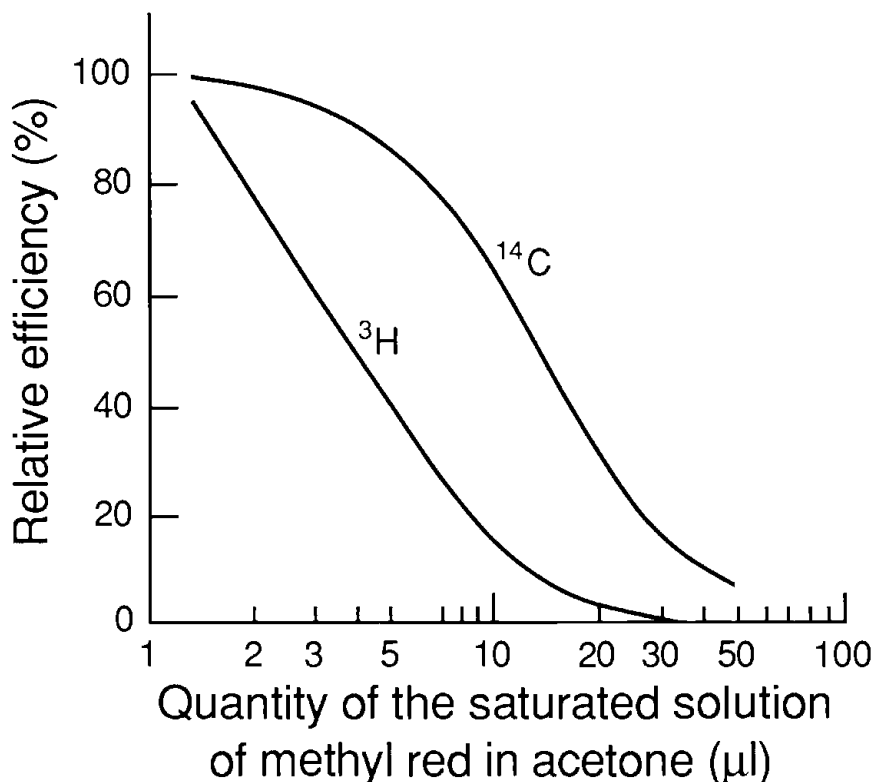
Figure 8. Chemical quench curves for <sup>3</sup>H and <sup>14</sup>C (quenching agents, acetone and carbon tetrachloride).

where  $E_{rc}$  and  $E_{rc1}$  are relative counting efficiencies caused by chemical quencher and color quencher in the mixed quench sample. The chemical quenching effect of methyl red was not considered.

The decrease in relative counting efficiencies of <sup>3</sup>H and <sup>14</sup>C with increasing amounts of carbon tetrachloride and acetone are shown in Figure 8. Likewise, changes in relative counting efficiencies due to the addition of increased quantities of saturated methyl red solution in acetone are shown in Figure 9. The equivalent relative counting efficiency of mixed quench samples can be represented by the product of the relative counting efficiencies of chemical and colour quenching in these samples (Table 2). In addition,  $F_c$  of chemical quenching can be calculated according to Equation 1 (Table 2). The quenching contribution factor for color quenching ( $F_{c1}$ ) equals  $1 - F_c$ .

Obviously, Equation 1 is usable only for the samples with known quenching components. When the quenching components are unknown, a new parameter, quenching type contribution factor (QTCF), has been defined by Yang<sup>4</sup> to represent the relative position of mixed quench samples in a mixed quench zone:

$$QTCF_{c1} = \frac{E_c - E_s}{E_c - E_{c1}} \quad QTCF_c = \frac{E_s - E_{c1}}{E_c - E_{c1}} \quad (2)$$

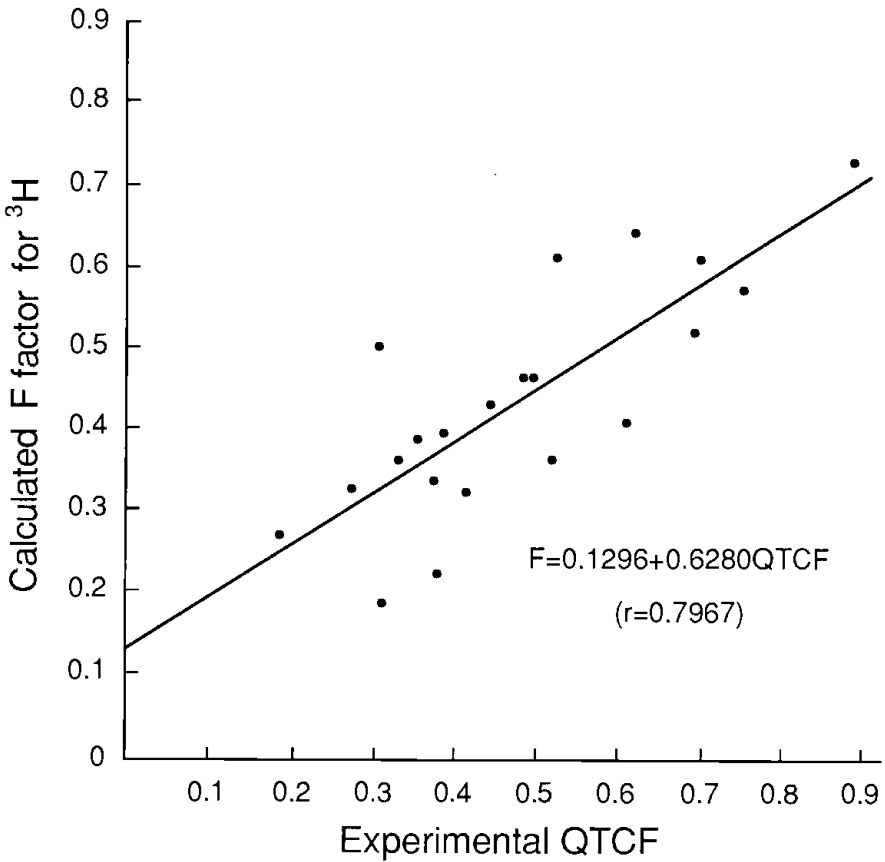


**Figure 9.** Color quench curves for  $^3\text{H}$  and  $^{14}\text{C}$  (quenching agent saturated solution of methyl red in acetone).

where  $E_c$  and  $E_{c1}$  are the counting efficiencies on the chemical and color quenching correction curve corresponding to the QIP (quenching indicating parameter) of the mixed quench sample.  $E_s$  is the counting efficiency of a mixed quench sample. In contrast, when QTCF is known we can calculate the counting efficiency of unknown mixed quench samples according to its QIP and QTCF:

$$\begin{aligned} E_s &= E_c - \text{QTCF}_{c1} (E_c - E_{c1}) \\ E_s &= E_{c1} + \text{QTCF}_c (E_c - E_{c1}) \end{aligned} \quad (3)$$

The QTCF value of the sample with unknown quenching components cannot be determined by Equation 2 as the counting efficiency  $E_s$  is unknown. The double vial method<sup>4</sup> has been used to solve this difficulty:  $1/m$  of total volume is taken from an unknown sample after the first counting and is diluted to original volume with the same cocktail, then it is recounted with the same conditions. Suppose  $\text{QIP}_1$  and  $\text{QIP}_2$  were obtained from the first and the second measurements, respectively; thus,



**Figure 10.** The relation of calculated F factor to experimental QTCF. These parameters were obtained from mixed quench samples.

$$\text{QTCF} = \frac{mrE_c'' - E_c'}{mr E_{cc}'' - E_{cc}'} \quad (4)$$

where  $E_c'$  and  $E_c''$  are the counting efficiencies from the chemical quenching correction curve corresponding to  $\text{QIP}_1$  and  $\text{QIP}_2$ , respectively. Therefore,  $E_{cc}' = E_c' - E_{c1}'$  and  $E_{cc}'' = E_c'' - E_{c1}''$ ; and  $E_{c1}'$  and  $E_{c1}''$  are the counting efficiencies on the color quenching correction curve corresponding to  $\text{QIP}_1$  and  $\text{QIP}_2$ , respectively; where  $r = n_2/n_1$ , and where  $n_1$  and  $n_2$  are the counting rates of the first and the second measurements, respectively. If the amount of the second sample was  $m$  times that of the first sample, then

$$\text{QTCF} = \frac{mE_c'' - rE_c'}{m E_{cc}'' - r E_{cc}'} \quad (5)$$

The reason for using the double vial method to determine QTCF is that the position of an unknown sample in a mixed quench zone cannot be determined by a single count, as in the usual quench correction method. Now, in this experiment the difficulty was resolved by the addition of  $^{67}\text{Ga}$  to the unknown sample. The quench correction curves mentioned above were replaced by double spectral index plots. The point distributive character of mixed quench samples in the mixed quench zone of double spectral index plots are the same as in quench correction curves. Therefore, QTCF can also be used, for example, when an unknown sample was measured and its SIE was determined. The chemical and color quenched  $\text{SIS}_s$  values in the energy range of 15 to 100 keV, corresponding to SIE in Figure 5, were  $\text{SIS}_c$  and  $\text{SIS}_{cl}$  respectively, while the SIS value of the sample itself was  $\text{SIS}_s$ . We can then obtain experimental QTCF values from Formulas 6 and 7 listed below:

$$\text{QTCF}_{cl} = \frac{\text{SIS}_c - \text{SIS}_s}{\text{SIS}_c - \text{SIS}_{cl}} \quad (6)$$

$$\text{QTCF}_c = \frac{\text{SIS}_s - \text{SIS}_{cl}}{\text{SIS}_c - \text{SIS}_{cl}} \quad (7)$$

where  $\text{SIS}_c - \text{SIS}_{cl}$ ,  $\text{SIS}_s - \text{SIS}_{cl}$ , and  $\text{SIS}_c - \text{SIS}_s$  are negative values because the color quenched curve was above the chemical quenched curve. However, the value of QTCF is always positive.  $\text{QTCF}_{cl}$  and  $\text{QTCF}_c$  characterize the degree of the contribution of color and chemical quench to mixed quench samples, respectively. The values of QTCF taken from Figure 7 are always the same as those taken from Figure 5, as only  $^{67}\text{Ga}$   $\text{SIS}_c$  and  $\text{SIS}_{cl}$  in the region of interest of 15 to 100 keV (corresponding to the SIE of a mixed quench sample) will change correspondingly to  $^{67}\text{Ga}$   $\text{SIS}_s$  in the region of interest of 0 to 7 keV. Thus, when we substitute QTCF into Equation 3, the counting efficiency of an unknown sample is resolved. The  $^3\text{H}$  corrected results, which are obtained by using QTCF (from Figure 5) and SIS quench correction curves, are given in Table 3. Except for one sample, the QTCF method gave more accurate values than those obtained by using either the chemical or color quenching correction curve alone. The mixed quench zone is wider in the range of strong quenching; therefore, the employment of QTCF for quench correction in that range has particularly significance.

## DISCUSSION

The experimental QTCF values of 21 mixed quench samples were obtained from Figure 3 and Equation 7 (Table 2). The correlation of the calculated F factor with experimental QTCF values for mixed quench samples was given in Fig. 10, in which  $F = 0.1296 + 0.6280 \text{ QTCF}$  ( $r = 0.7967$ ). It is illustrated that F factor is proportionally related to QTCF; in fact, this is the basis of the

Table 3. The Comparison Between Activities Obtained Using QTCF and Activities Obtained Using the Usual Quench Correcting Curves for Strongly Mixed Quench Samples<sup>a</sup>

Sample Number	SIS of 0-19 keV	Sample Counts (cpm)	Counting Efficiency			Activity (dpm)			Using QTCF	Error (%) <sup>b</sup>	
			Chem. (%) <sup>c</sup>	Color (%) <sup>c</sup>	QTCE (%) <sup>d</sup>	Chem. Curve	Error (%) <sup>b</sup>	Color Curve			Error (%) <sup>b</sup>
1	12.8	6290	24.5	20.4	22.9	25673	-14.5	30833	2.7	27467	-8.5
2	12.4	5610	21.9	17.4	18.8	25616	-14.6	32241	7.4	29840	-0.6
3	11.2	3204	13.2	9.1	10.2	24273	-19.1	35209	17.3	31412	4.7
4	12.1	5328	20.0	15.3	18.9	26640	-11.2	34824	16.0	28190	-6.1
5	11.1	2917	12.6	8.5	10.6	23151	-22.9	34318	14.3	27519	-8.3
6	11.8	4444	17.8	13.2	15.6	24966	-16.8	33667	12.2	28487	-5.1
7	10.8	2445	10.7	6.3	8.3	22850	-23.9	38810	29.3	29458	-1.9
							-17.6 ± 4.3		14.2 ± 7.8		-3.7 ± 4.4

<sup>a</sup>Each sample contained 20  $\mu\text{L}$  aqueous solution of  $^3\text{H}$ -thymidine (30014 dpm).

<sup>b</sup>Error (%) =  $\frac{\text{actual activity} - \text{corrected activity}}{\text{actual activity}} \times 100$

<sup>c</sup>Taken from curves in Figure 7 (left)

<sup>d</sup>Counting efficiencies were obtained by QTCF from Figure 3.

quench correction method for mixed quench samples in this chapter. If we call the usual quench correction "first order quench correction," then QTFC quench correction in mixed quench zone can be called "second order quench correction." Obviously, the magnitude of the second order quench correction is less than the first order quench correction; therefore, it must have greater relative variability. Based on this point, in principle we can divide the mixed quench zone into subzones by the measurement of two parameters of the curve. All points falling in the same subzone were considered to have the same QTCF value which is more practical for application.

If a liquid scintillation counting system can automatically give two spectral indexes for two different parts of a sample (or external standard) spectrum at the same time, it is not only convenient for measurement of double spectral index plots, but also more usable to various new and existing applications. The authors had hoped to prepare double spectral index plots of external standards to correct mixed quench samples, but the commercial instrument used was unable to measure the SIE of different energy ranges.

The main advantages of using  $^{67}\text{Ga}$  or  $^{113\text{m}}\text{In}$  as an internal standard include, (1) the elimination of accurate pipetting of the internal standard, (2) the ability to recover the sample after the spiked activity has decayed, and (3) the high count rate.<sup>11</sup> Among these advantages the one with the greatest importance is probably the fact that the content of "standard" was changed from quantity of activity to energy or its spectrum shape. The mixed quench samples in this paper consisted of simple chemical and color quenchers. Practical samples are very complex, therefore, it is necessary to do further work. In addition, further work with the double spectral index plots or ratio of spectral index may yield some new applications.

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