

# 5 Liquid Scintillation Counting

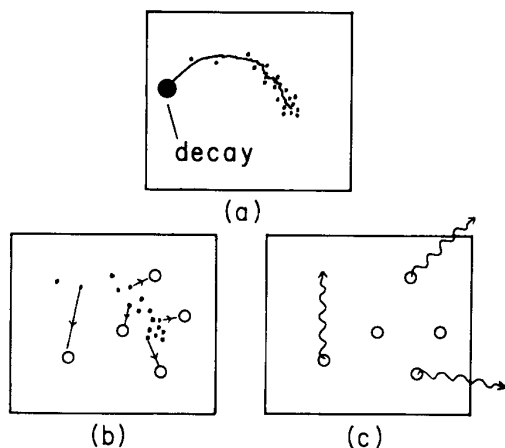
## 5.1 The weak radiation problem

Carbon-14 and tritium are  $\beta^-$  emitting radioisotopes with very low energy  $\beta^-$  emissions which are extremely difficult to detect with any form of window counter, due to self-absorption of the  $\beta^-$  particles and their absorption within the counter window. The window problem has been overcome to some extent by counting solid samples within a windowless G-M or proportional counter, where efficiencies of 15% for  $^{14}\text{C}$  and 2.5% for  $^3\text{H}$  have been reported. To reduce the self-absorption losses it is desirable to mix the active sample homogeneously with the detecting material. This can be done by counting the sample in the gaseous phase. For example,  $^{14}\text{C}$  samples may be converted into  $^{14}\text{CO}_2$  by combustion or by evolution from a labelled carbonate. The gaseous activity can then be intimately mixed with the filling gas of any type of gas ionisation detector, thus minimising the effect of  $\beta^-$  absorption and resulting in high counting efficiencies. Tritium can also be counted in the gaseous state as hydrogen, water vapour or a hydrocarbon.

Since the 1960s a second method of overcoming the  $\beta^-$  absorption problem has become widely used. In this the radioactive sample and a scintillator material are both dissolved in a suitable solvent, and the resulting scintillations are detected and counted. The method is called liquid scintillation counting.

## 5.2 Outline of liquid scintillation counting

If a compound containing an  $\alpha$  or  $\beta$  emitting isotope is dissolved in a solvent such as toluene, the radioactive emissions result in the formation of electronically excited solvent molecules. If the solution also contains a small amount of a suitable scintillator, the excited solvent molecules rapidly transfer their excitation energy to the scintillator, forming electronically excited scintillator molecules, which then relax by the emission of photons. The processes involved are summarised in *Figure 5.1*.

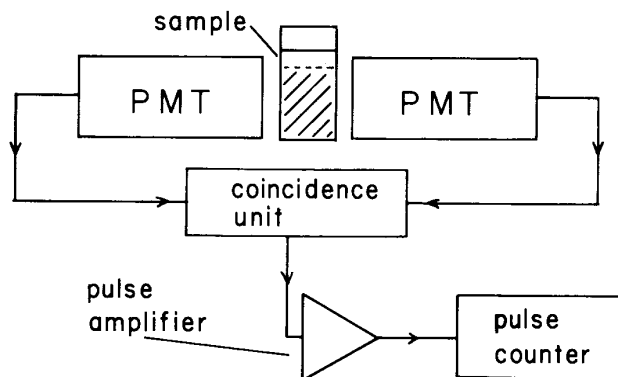


*Figure 5.1 The mechanism of visible light photon emission which forms the basis of liquid scintillation counting. (a) Passage of radiation through the solution produces a trail of ionised and electronically excited solvent molecules (•). (b) Excited solvent molecules transfer their energy to solute molecules producing electronically excited solute molecules (o). (c) Electronically excited solute*

*molecules lose their excitation energy by collisional de-excitation or by photon emission*

The scintillator material is usually chosen so that the wavelength of the emitted light is in a region of the spectrum which can be conveniently detected by a photomultiplier tube, e.g. blue light. The result of the combination of radioisotope, solvent and scintillator is that each radioactive decay in the solution gives rise to a flash of visible light, so that electronic counting of these scintillations gives a measure of the activity of the radioactive material. As we shall see later, one of the problems in liquid scintillation counting is that in fact not all the scintillations can be detected, so that the counting efficiency is normally  $< 100\%$  and the actual efficiency must be determined before the activity of the radioactive material can be calculated.

While it is possible to arrange for the scintillation to be detected using a single photomultiplier tube, the general noise level of a PMT at room temperature is so high that the separation of the signal pulses (due to scintillations) from the noise pulses (due to thermionic emission from the photocathode of the PMT) becomes difficult. This is especially true for low decay energy isotopes as the  $\beta$  emitter  $^3\text{H}$ ,  $^{14}\text{C}$  and  $^{35}\text{S}$ . Cooling the PMT can help reduce thermionic noise but in fact most commercial liquid scintillation counters use two PMTs, recording a count only when a scintillation is detected by both tubes within a short time period (usually  $1\mu\text{s}$ ). This arrangement is shown in *Figure 5.2*. The technique is called coincidence counting and is highly effective at discriminating against the random noise pulses from the two tubes.



*Figure 5.2 Schematic arrangement of photomultiplier tubes (PMTs) and associated electronic units required for coincidence counting in simple liquid scintillation counters. The PMTs and the sample are housed in a light-proof enclosure*

The counting efficiency of a liquid scintillation system may be defined as

$$\frac{\text{Number of scintillations per minute detected}}{\text{Number of disintegrations per minute in solution}}$$

Clearly this overall efficiency is made up of two components: one from the scintillator solution itself, and one from the photon detection system and its associated electronics. We will consider these two aspects of the system in turn.

### **5.3 Liquid scintillator solutions**

A wide range of scintillator solutions is available in modern radiochemical laboratories. To make a sensible choice for a particular application, the function of the components of the solution must be understood. All scintillator solutions contain:

(1) a solvent; (2) a primary 'solute' - the scintillator material, and may contain (3) a secondary solute. The nature and functions of each component are considered below.

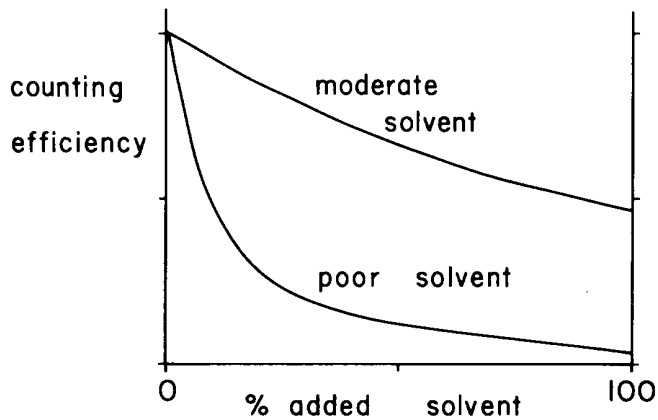
#### (1) The solvent

Despite its name the function of the solvent is not simply to dissolve the radioactive sample. The solvent's functions are to keep the scintillator or solute in solution, and to absorb the decay energy of the radioisotope for subsequent transfer to the solute. Changes to the solvent, such as dilution with other material, may have a marked effect on the efficiency with which the solvent fulfils these roles.

Solvents fall broadly into three categories:

- (i) Effective solvents, e.g. the aromatic hydrocarbons, of which toluene and xylene are by far the most widely used.
- (ii) Moderate solvents, e.g. many non-aromatic hydrocarbons. With appropriate scintillator these may result in counting efficiencies of 15-40% of that of toluene.
- (iii) Poor solvents; unfortunately this is virtually everything else including most common laboratory solvents such as alcohols, ketones, esters and chlorinated hydrocarbons. Poor solvents usually give solutions with counting efficiencies of 1% of that of toluene.

The effect of adding a moderate or poor solvent to a liquid scintillator solution based on an effective solvent is shown in *Figure 5.3*. The effect on the counting efficiency is interesting because most of the samples that one has occasion to count tend to come from the poor solvent category.

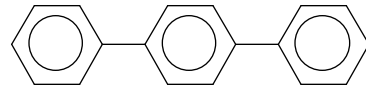


*Figure 5.3 Typical reduction of counting efficiency of a liquid scintillator solution as a poor or moderate solvent is added to the solution. The quantitative effect will depend on the nature of the radionuclide present and the scintillator solute in the solution*

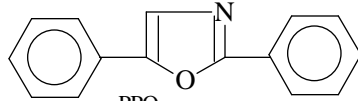
#### (2) The primary solute

This is the name given to the scintillator material in the solution. The solute acts as a trap for the radioactive decay energy initially converted into electronic excitation energy by the solvent molecules. The earliest solutes were naphthalene, anthracene and *p*-terphenyl, and large conjugated systems are still the most widely used, and indeed the most efficient. Modern scintillator solutes often have lengthy chemical

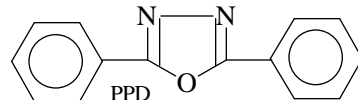
names and are most frequently known by simple abbreviations. Among the commonest are:



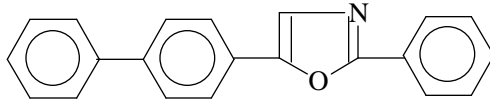
p-terphenyl



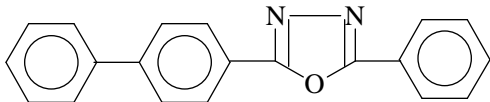
PPO  
1-phenyl, 4-phenyl oxazole



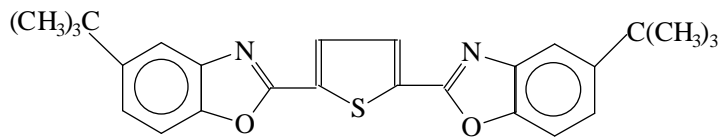
PPD  
1-phenyl, 4-phenyl oxadiazole



PBO 2-phenyl, 5(4-biphenyl) oxazole



BPD 2-phenyl, 5(4-biphenyl) oxadiazole



BBOT 2,5 di-(5-t-butyl-2-benzooxazolyl) thiophene

As the concentration of primary solute in a scintillator solution increases so the counting efficiency for a sample of, say,  $^{14}\text{C}$  initially increases, as shown in *Figure 5.4*. However, a scintillator solute, being an efficient photon emitter, is also a good photon absorber, so that a concentration is reached at which the number of photons escaping from the solution begins to fall. As a result there exists an optimum solute concentration at which maximum counting efficiency occurs. *Table 5.1* shows primary solute concentrations generally used in popular scintillator solutions.

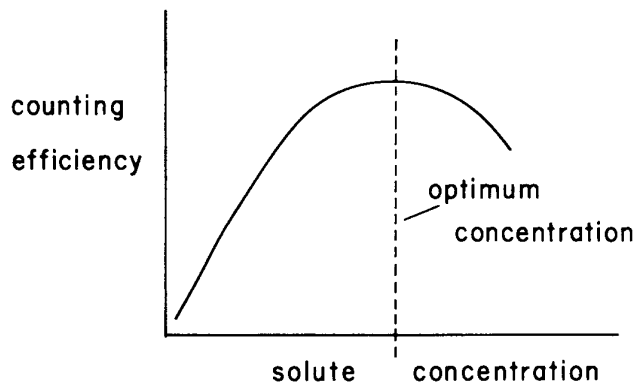


Figure 5.4 Typical variation of counting efficiency of a liquid scintillator solution with the concentration of the primary solute. The actual variation will depend on the nature of the radionuclide present and on the nature of any other compounds in the solution

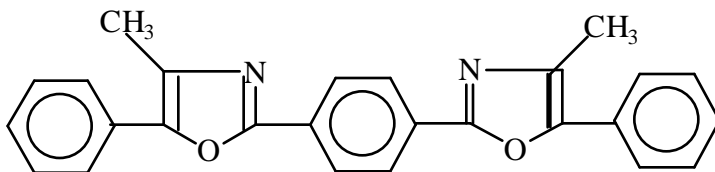
Table 5.1 Primary solute concentrations of popular scintillator solutions

Solute	Concentration (g l <sup>-1</sup> )
p-terphenyl	5
PPO	3-7
PBD	8-10
BBOT	7

### (3) The secondary solute

The photons emitted by PPO - probably the most popular primary solute - have wavelengths in the range 300-400 nm, a region of the spectrum which is ideal for detection by modern photomultiplier tubes. Unfortunately a large number of molecules have inconvenient photon absorptions in this region of the spectrum, particularly molecules of interest in biochemistry and medicinal chemistry, and samples containing such molecules would be counted with a lowered counting efficiency. When high counting efficiency is important the absorption of photons by either the sample or the primary solute may be reduced by using a secondary solute, which traps the excitation energy from the primary solute and emits photons of a longer wavelength.

The most important secondary solutes are POPOP and its dimethyl derivative. Dimethyl POPOP is 1,4-di-2-(4-methyl, 5-phenyl oxazolyl) benzene



and emits photons in the 400-500 nm range. Both materials have rather low solubilities in common scintillator solvents and are normally used in concentrations of 0.05-0.2 g l<sup>-1</sup>

The combination of solvent and solutes, together with additives which are sometimes used to improve miscibility of the sample material with the solvent, is

referred to as a scintillator cocktail. Although an excellent range of scintillator cocktails is available from commercial suppliers, some laboratories find it worthwhile to mix their own and a selection of commonly used recipes is given in *Table 5.2*. Note that purified materials, including the solvent, should be used to make these cocktails if problems such as chemiluminescent photon emission are to be avoided. 'Scintillation grade' solvents and solutes are available from many suppliers, and the slightly higher cost than the laboratory reagent grade is usually worthwhile.

Table 5.2 Typical scintillator cocktails (concentrations in  $\text{g l}^{-1}$ )

<i>Solvent</i>	<i>Primary solute</i>	<i>Secondary solute</i>	<i>Additives</i>	<i>Sample type</i>
Toluene	PPO (4-6)	Me <sub>2</sub> POPOP (0.05-0.2)	--	organic soluble
Toluene	butyl PBD (8-12)	- --	-	organic soluble
p-Xylene	butyl PBD (8-2)		Ethanol (100)	aqueous
1,4 Dioxan	PPO (4-6)	Me <sub>2</sub> POPOP (0.2)	Methanol (100) Ethylene glycol (20) Naphthalene (60)	aqueous

#### 5.4 Sample preparation

There are many methods of preparing a sample for liquid scintillation counting. In each case the aim is to achieve a mix between sample and scintillator which will ensure first that the decay radiation deposits its energy within the scintillator solution, and second that the photons emitted during scintillation can escape from the sample bottle and be detected by the photomultiplier tubes. A few of the available methods are described briefly below.

##### (1) Direct solubilisation

If the radioactive sample happens to be soluble in the scintillator solvent (for example, toluene for organic soluble samples or dioxan for aqueous samples) then a small amount of sample can be dissolved directly in the scintillator cocktail. While this is the simplest method of sample preparation there can be difficulties. For example, when aqueous samples are mixed with a cocktail one occasionally observes the precipitation of the primary solute, a problem which may be exacerbated by refrigeration of the mixture. Many samples can be rendered soluble by a relatively straightforward chemical process, such as complexing with alkyl phosphoric acids, a procedure which has been used for holding radioisotopes of metal ions in scintillator solutions.

##### (2) Action of a solubiliser

Tissues, proteins, nucleic acids and a wide range of macromolecules may often be converted into a soluble form using a solubiliser. Quaternary ammonium salts are useful solubilisers, although it should be noted that chemiluminescent processes can occur unless highly purified material is used. Some companies supply a 'scintillation grade' range of these salts. Other commercial solubilisers are marketed under names such as Hyamine Hydroxide, Soluene, Protosol and Tissue Solubiliser. Some liquid

scintillation cocktails are supplied with a solubiliser already added so that macromolecular materials can be dissolved directly.

### (3) Gel counting

While solid insoluble samples may be counted on filter papers carefully positioned within the sample bottle, the result is usually poor in efficiency and reproducibility. It is generally better to count the sample as a fine powder suspended in the scintillator cocktail. Many powders will remain in suspension if broken up using ultrasonics. For dense particles the viscosity of the cocktail may be increased to prevent the settling out of the sample before counting is completed. Thickening agents available for this purpose include aluminium stearate, toluene di-isocyanate (which is carcinogenic) and several polyolefinic resins. In some cases the mixture must be heated to induce thickening.  $\alpha$  and low energy  $\beta^-$  emitting isotopes may be difficult to count reproducibly by this method unless the particle size can be controlled, as some self-absorption of the radiation may occur. For the same reason determination of the absolute counting efficiency may be difficult.

### (4) Emulsion counting

Liquid samples (usually aqueous solutions) which are not miscible with an aromatic based scintillator cocktail can be dispersed to form an emulsion. As with solid particles in gel counting, the size of the micelles is very important in determining the counting efficiency for low energy emitters, and again ultrasonics may be used to obtain small micelles. Emulsions of this type are generally stabilised by using an emulsifier-typically a polyethoxylated surfactant. Commercially available emulsifiers such as Triton N.104 are excellent, although there have been reports that much cheaper industrial detergents can be just as good. Several ready-made emulsion liquid scintillator cocktails are commercially available, and some can accept quite large quantities of sample. For example, a good micellar scintillator, can accept up to 40% water. Many of these cocktails can be used for counting a wide range of water-soluble proteins, nucleotides, salts and sugars.

Most samples for liquid scintillation counting are counted in small vials which hold 15-20 ml, although in recent years liquid scintillation counters have become available for counting minivials holding about 5 ml of liquid. Sample vials are usually made of polythene or glass. Polythene vials are disposable and cheap, but cannot be used for holding some organic solvents for long periods. Although they may not look transparent, in fact they transmit the scintillation photons slightly more efficiently than clear glass. Glass bottles allow easier inspection of the sample and can be used to store samples for moderate periods. Ordinary glass bottles can be troublesome for very low activity counting because of the presence of the radioisotope  $^{40}\text{K}$  in the glass. Low potassium vials are available from many suppliers, although they are more expensive than ordinary glass bottles. Some laboratories re-use glass sample vials after washing and checking for contamination.

Samples for liquid scintillation counting must be moved around the laboratory (from workbench to counter, and so on) and I prefer to use polythene sample bottles where possible, simply because they bounce when dropped.

## 5.5 Counting channels

Liquid scintillation counting is most frequently used for counting  $\beta^-$  emitting isotopes and it has been found that the number of photons emitted per  $\beta^-$  decay (and hence the pulse height) is proportional to the energy of the  $\beta^-$  particle. Thus with a suitable photon detection system a liquid scintillation counter operates as a  $\beta^-$  energy spectrometer. The average energies of  $\beta^-$  particles emitted by  $^3\text{H}$ ,  $^{14}\text{C}$  and  $^{32}\text{P}$  (three of

the most commonly used radioisotopes) are ~ 5 keV, 50 keV and 500 keV respectively. To obtain countable electronic pulses from photomultiplier tubes operating under fixed conditions, and viewing scintillations which vary in intensity by more than three orders of magnitude, it is common practice to follow the photomultiplier tubes with either a logarithmic amplifier or several separated linear amplifiers in parallel (each with a gain optimised for one particular range of  $\beta$  energies).

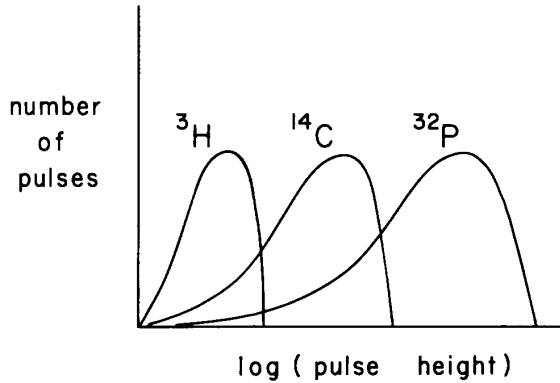


Figure 5.5 Pulse height distributions obtained during decay of  $^3\text{H}$ ,  $^{14}\text{C}$  and  $^{32}\text{P}$  in a liquid scintillation counter

With a logarithmic amplifier the amplified pulse heights produced by the  $\beta$  decays of the three radioisotopes have distributions as shown in Figure 5.5. Using an upper and lower level pulse height discriminator it is easy to select a range of pulse heights (equivalent to a range of  $\beta$  decay energies) for counting, so that pulses from one radionuclide are counted with high efficiency while pulses from noise or other isotopes are essentially ignored. For example, with the discriminators set as shown in Figure 5.6 the  $^{14}\text{C}$  isotope is counted while decays of  $^3\text{H}$  or  $^{32}\text{P}$  are largely ignored. The range of pulse height accepted by the counter is that which lies in the window or channel between the two discriminators. Clearly the channel shown in Figure 5.6 would allow  $^{14}\text{C}$  to be counted in the presence of  $^3\text{H}$  or  $^{32}\text{P}$  without major interference from the latter isotopes.

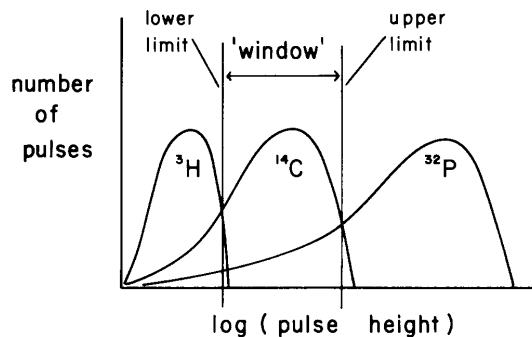


Figure 5.6 Positioning of pulse height limits to set up a window or channel suitable for counting  $^{14}\text{C}$  decays without major interference from  $^3\text{H}$  or  $^{32}\text{P}$

Most commercial liquid scintillation counters have facilities for switching to lower and upper level discriminators which have been preset as  $^3\text{H}$ ,  $^{14}\text{C}$  or  $^{32}\text{P}$  channels, so that these isotopes may be efficiently counted either alone or in the presence of one another. A similar arrangement is adopted in liquid scintillation



counters with several separate linear amplifiers and in the latest generation of microprocessor controlled/digital memory instruments available commercially. Many modern instruments also have the ability to count pulse heights in several channels simultaneously, so that two or more radioisotopes may be counted independently in a single sample and at the same time.

Modern photomultipliers and electronic instrumentation have resulted in liquid scintillation counters which can detect even the low energy  $\beta^-$  emissions of  $^3\text{H}$  with high efficiency. Although the efficiency depends on the channel width used for counting and the noise level of the system, modern instruments may count  $^3\text{H}$  samples with efficiencies up to about 50%.  $^{14}\text{C}$  and  $^{35}\text{S}$  samples may be counted with up to 95% efficiency, and most high energy  $\beta^-$  emitters (e.g.  $^{32}\text{P}$  with up to 99% efficiency). In practice the counting efficiencies are often lower than these values because of the presence of impurities, sample solvents and other materials from the 'poor solvent' category. This reduction of counting efficiency is called quenching.

Quenching reduces the number of photons emitted from a scintillator solution for a given radioactive decay energy. The magnitude of the electrical pulse reported by the photomultiplier tubes is reduced as a result, so the efficiency of counting for a particular radioisotope varies with the degree of quenching within the solution. Quenching effects in fact fall into two categories: (i) chemical quenching, caused by de-excitation of electronically excited molecules which would otherwise give rise to emitted photons; and (ii) colour quenching, caused by absorption of emitted photons by materials in the solution usually a coloured sample. While (ii) may sometimes be overcome by the use of a secondary solute such as POPOP, which moves the wavelength of the emitted photons to a different region of the spectrum, there is usually little that can be done about (i) except to restrict the amount of quenching agent added to the scintillator cocktail.

For the low energy  $\beta^-$  emitting isotopes, particularly  $^3\text{H}$ , quenching is often severe. Oxygen gas dissolved in a scintillator cocktail can reduce counting efficiency by 20% for a  $^3\text{H}$  sample and for this reason scintillator cocktails are often flushed with nitrogen or argon before use. Samples stored in refrigerators are particularly prone to contamination by water during sample preparation, again causing significant quenching problems. In general there are few occasions on which one can afford to ignore the effect of quenching on the counting efficiency in liquid scintillation counting, and it is usual practice to determine the counting efficiency with which every sample is counted.

## 5.6 Estimation of counting efficiency

There are a number of ways of estimating counting efficiency in liquid scintillation counting. Fortunately the methods employed on most commercial instruments are variations on the two basic techniques described below. As there is a sense in which all lowering of counting efficiency below 100% is a result of quenching, the procedure of estimating the efficiency of counting is frequently referred to as the estimation of the quench correction.

### (1) The sample channels' ratio method (SCR)

Because all liquid scintillation counters have facilities for setting channels for particular isotopes, any individual isotope may be counted in two separate channels as illustrated in *Figure 5.7* for an unquenched  $^{14}\text{C}$  sample. In this example channel A is a standard  $^{14}\text{C}$  channel and the counts recorded in this channel are counts required for the sample activity measurement. A second channel, B, can be set to record about one tenth of the  $\beta$  decays counted in channel A.

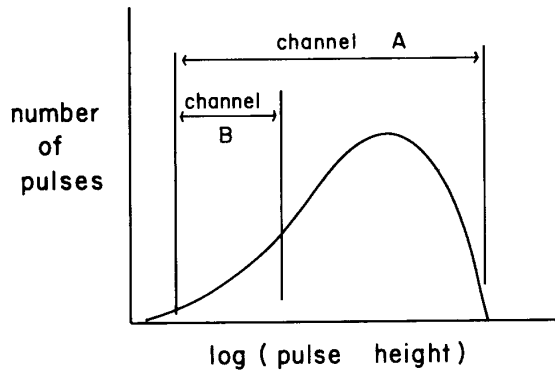


Figure 5.7 Two channels set up so that  $^{14}\text{C}$  may be counted in channel A and a sample channels ratio may be obtained using the ratio A/B

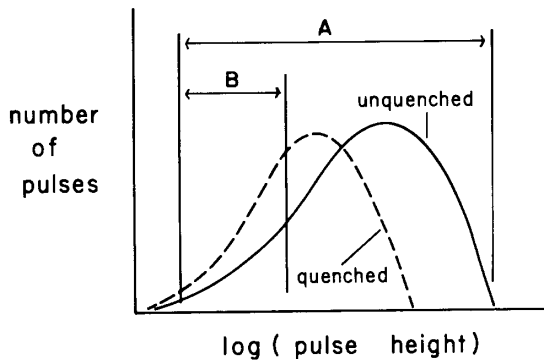


Figure 5.8 Effects of quenching on the position of the pulse height distribution. Note that quenching results in a greater portion of the distribution being counted in channel B thus lowering the ratio A/B

When quenching occurs the magnitude of every pulse is affected, so that the whole pulse height spectrum produced by the  $\beta^-$  decays moves to lower pulse heights, as shown in Figure 5.8. Clearly the ratio of counts recorded in the two channels, A/B, is different in the quenched solution from that in the unquenched solution ( $\sim 10$  in the unquenched spectrum and  $\sim 3$  in the quenched spectrum of Figure 5.8). In practice a calibration graph of absolute counting efficiency in channel A versus the channels ratio, A/B, is drawn using standards of known activity and varying degrees of quenching. A typical 'quench correction curve' of this type is shown in Figure 5.9. Once the calibration curve has been drawn, subsequent samples are simply counted in the two channels A and B, the channels ratio calculated and the counting efficiency in channel A read directly from the calibration graph.

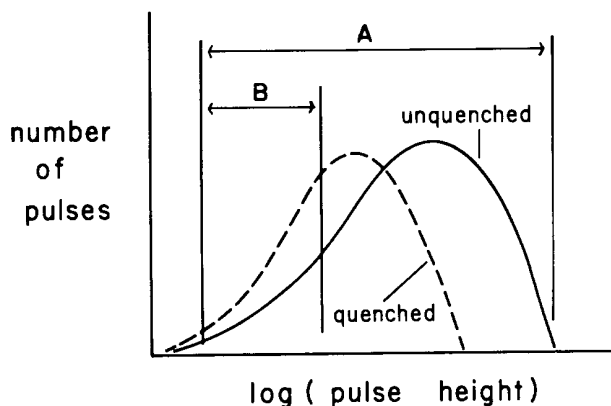


Figure 5.9 Typical calibration curve for efficiency estimation often called a quench correction curve

Most liquid scintillation counter suppliers will supply series of quench samples of radioactive material in scintillator cocktail (usually 10 or 12 samples of identical activity but differing degrees of quenching) so that the quench correction curves there must clearly be a different curve for each isotope and each counting channel may be checked or redrawn at regular intervals.

The disadvantage of the sample channels ratio method of efficiency estimation is that for low activity samples the time required to record a statistically useful number of counts in the smaller channel (channel B in Figure 5.8) may be long. This problem is largely overcome by the second method of quench correction.

## (2) The external standard channels' ratio method (ESCR)

The ESCR method is based on the same reasoning as the SCR method. The difference between the two is that for the ESCR method the pulse spectrum used for the ratio measurement - not, of course, for the sample activity measurement - is provided by electrons scattered within the scintillator solution by an external  $\gamma$  photon source (usually  $^{137}\text{Cs}$  or  $^{226}\text{Ra}$ ). Clearly this approach requires three separate measurements: one of the sample in the appropriate isotope channel; one of the sample in the two preset ESCR channels, and one of the counts produced by the source in the two ESCR channels, the second set of counts being subtracted from the third before the ratio is calculated. Both sets of counts in the ESCR channels may be performed quite rapidly since a high activity  $\gamma$  source (typically several hundred kBq) may be used.

As before, a calibration curve of counting efficiency versus channels ratio is drawn using quenched standards of known activity, and this is used to determine the absolute counting efficiency of samples on a routine basis.

Whichever method of quench correction is used, clearly there must be a different calibration curve for each isotope to be counted. Furthermore both methods, while usually quite reliable for chemical quenching, must be used cautiously for colour quenching, because in the latter the scintillation photons may be selectively absorbed and the shape of the pulse height spectrum modified. Certainly for accurate work a separate quench calibration curve should be produced for coloured samples.

Many of the new generation of liquid scintillation counters contain a computer and may be programmed to perform some kind of efficiency estimation, in some cases converting the recorded sample count into the estimated absolute activity of the sample complete with appropriate error limits on the reported value. The principal advantage of liquid scintillation counting, i.e. the lack of a barrier between the

radioactive sample and the detecting device, has resulted in the use of liquid scintillation counters for counting virtually all  $\alpha$  and  $\beta^-$  decay isotopes. Scintillator cocktails containing heavy-metal compounds are also available, and these can be used for counting  $\gamma$ -emitting nuclides - particularly low energy  $\gamma$  emitters such as  $^{125}\text{I}$ . In fact it is probably true to say that the single most useful item of counting equipment for any radiochemical laboratory is a liquid scintillation counter, as there are very few radionuclides which cannot be counted with an acceptable efficiency in such an instrument.

### 5.7 Cerenkov counting

While virtually any radionuclide can be counted using liquid scintillation counting, problems can arise in the sample preparation stage simply because an organic scintillator material needs to be in intimate contact with the radioactive sample. While the use of gel or emulsion counting may often overcome problems of incompatibility, there is an alternative and very useful technique for counting energetic  $\beta^+$  and  $\beta^-$  decay isotopes which does not involve the use of a scintillator material at all. The method is based on the Cerenkov effect, which is the emission of a bluish-white light (Cerenkov radiation) when an electron or positron travels through a medium with a velocity which is greater than the velocity of light in that medium. (The velocity of light in a vacuum,  $c$ , cannot be exceeded: light travels more slowly through matter.)

The minimum particle velocity, and hence the threshold energy, at which the Cerenkov effect is observed depends on the refractive index of the medium involved. In most common solvents the Cerenkov threshold for  $\beta^-$  particles lies between 0.15 and 0.27 MeV. In water, for example, the minimum  $\beta^-$  particle energy which results in the emission of Cerenkov radiation is 0.263 MeV. As energetic  $\beta^-$  particles pass through matter emitting Cerenkov radiation they lose energy, primarily by ionisation and electronic excitation processes, and quickly fall below the Cerenkov threshold. Consequently the Cerenkov radiation emitted by  $\beta^-$  particles in a solution of a radioisotope appear as brief flashes of light, which may be detected and counted in much the same way as the scintillations in liquid scintillation counting.

For energetic  $\beta^-$  emitters such as  $^{24}\text{Na}$  and  $^{32}\text{P}$ , most of the  $\beta^-$  particles emitted are above the Cerenkov threshold for water (84% and 90% respectively), so that these isotopes may be detected and counted as aqueous solutions simply by placing a vial of the aqueous solution in a liquid scintillation counter. As no scintillator cocktail is required, larger sample volumes can be accommodated in the standard vial than would be possible in conventional liquid scintillation counting.

Unless one does a large amount of Cerenkov counting it is usually most convenient to count Cerenkov radiation in the 3 H channel of a liquid scintillation counter-this being the channel covering the lowest pulse height region. Of course, the counting efficiencies for different isotopes in this channel must be determined experimentally, but a rough guide to the efficiencies to be expected in water are given in *Table 5.3*. It should be noted that the manner in which Cerenkov radiation is emitted results in the efficiency of detection in a liquid scintillation counter being a function of sample volume, the optimum volume in many instruments being about one half the volume of a standard liquid scintillation vial. Hence all samples to be counted by Cerenkov counting should be of the same volume, additional solvent being added to some samples if necessary. It is clear from *Table 5.3* that counting efficiencies attainable by Cerenkov counting are not as high as would be expected using liquid scintillation counting. Nevertheless, the great simplicity of sample preparation may often go some way to compensating for this.

Table 5.3 Typical Cerenkov counting efficiencies for various radionuclides in  $^3\text{H}$  channel of liquid scintillation counters

<i>Radionuclide</i>	<i>Cerenkov counting efficiency * (%)</i>
37Cs	2
36Cl	2
47Ca	7
40K	14
24Na	18
32P	

\*Samples in aqueous solution.

Solvents other than water are also used for Cerenkov counting. Saturated hydrocarbons, halocarbons and alcohols are frequently useful, but if unsaturated or aromatic solvents are to be involved one must take particular care to check the counting efficiency, as a weak fluorescence from the solvent may boost the counting efficiency dramatically. While chemical quenching does not occur in Cerenkov counting, colour quenching can be a problem for coloured samples. Bleaching can often solve the problem, although where this is not practical the colour quenching can be accounted for using techniques which are analogous to the channels' ratio methods of liquid scintillation counting.

As the counting efficiency is largely unaffected by the chemical content of the sample solution, Cerenkov counting is often used for counting materials that have been digested by rather drastic chemical means. For example, labelled biological material may have been treated with perchloric acid before counting. Note that low level chemiluminescence is a fairly widespread phenomenon and may produce misleading count rates from systems which are not obviously emitting light. Control samples, from which the radioisotopes have been omitted, provide a useful check for such problems.

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