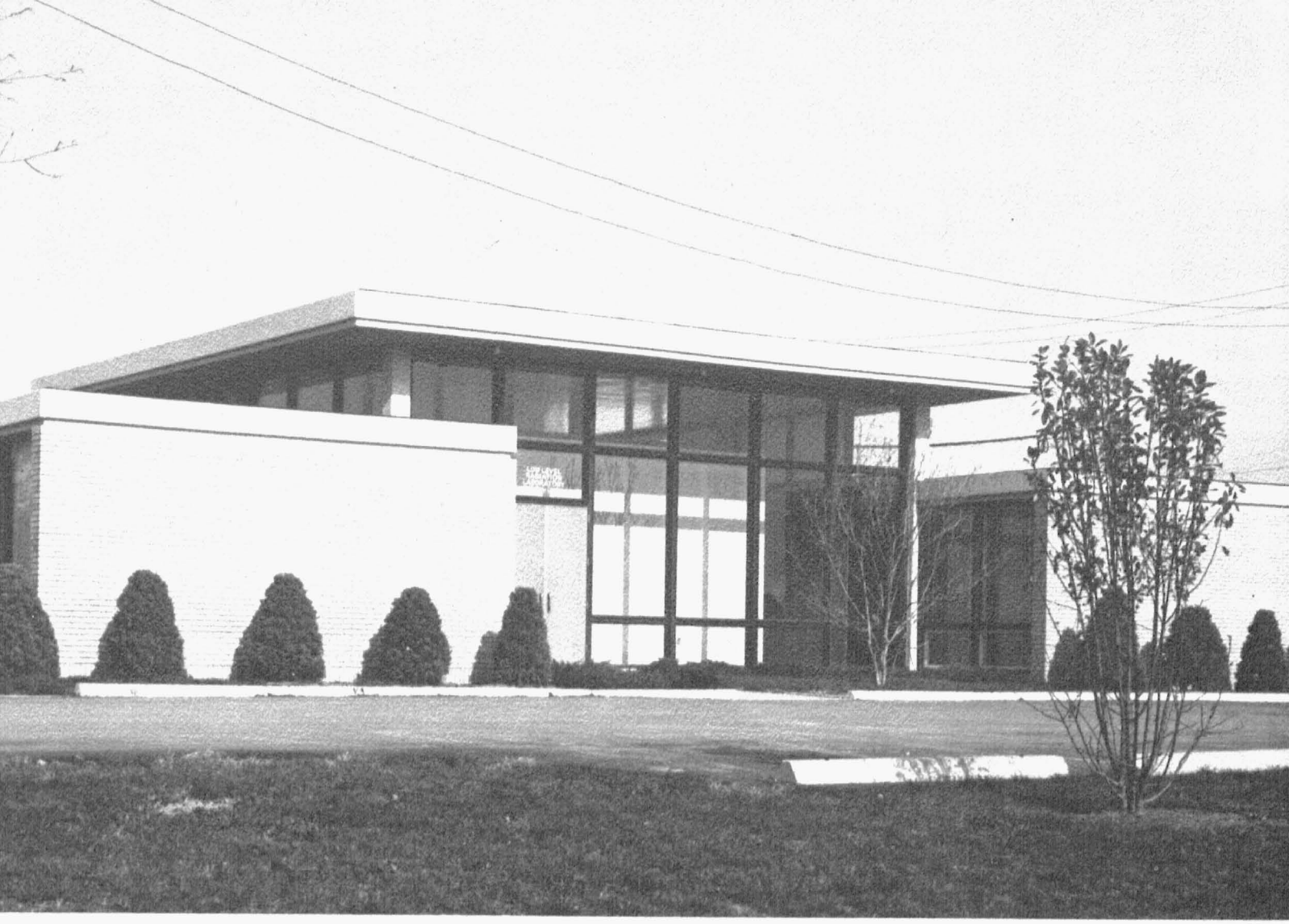

Manual for Use of

Two-Pi Liquid Scintillation Whole Body Counter

- ^{40}K Measurements
- Bio-Applications
- Counting Statistics

SR 88 APRIL, 1968
AGRICULTURAL EXPERIMENT STATION
UNIVERSITY OF MISSOURI - COLUMBIA

Preface

Dubos of Rockefeller University recently expressed the view that some investigators are prone to the *cult of serendipity,* which he describes as the equivalent of Stephen Vincent Benet's line, "We don't know where we are going, but we're on our way" (1).

This term provides an appropriate description of some who, during the early developmental stages of familiarizing themselves with new equipment, approach the task with an attitude of, "Let's see what it will do." Whether the term is meritorious or not depends on one's scientific philosophy and is not of question here. A point of importance, however, is that a great deal of information is gained by this approach which eventually will lead to a more detailed and disciplined study of the calibration, standardization, and control of the instrument.

Whole body counting is still in an infant stage of development. The accumulation of technical information is slow to come by. No one suspected the amount of "down time" which was to be experienced during the first year of operation; but some of the materials derived could not have been obtained by any other means.

The information contained in this manual has been acquired over an 18 month period. Originally I had planned to write a brief description of the counter and the technique used for standardizing and balancing. It was soon apparent that additional information should be included for teaching and research interests.

The section on bioapplications, although not original, provides a different approach for analyzing data. I hope it will be of some benefit to the student who is beginning to develop an interest in radiobiology.

I would like to acknowledge the helpful comments and suggestions of Doctors Ralph Anderson, James Ticer, Joyce Patterson and Mohammond Yousef in reviewing this manuscript. Major credit for establishment of the whole body counter at the University of Missouri is due to Dr. Ellis R. Graham and Dr. Steve E. Zobrisky. Without their continued encouragement and interest the dream might not have become a reality. They have also been a stimulus to my continued enthusiasm for this project. Vern Walter and Ben Schmidtke of Packard Instrument Co. provided valuable technical consultation.

William J. Coffman

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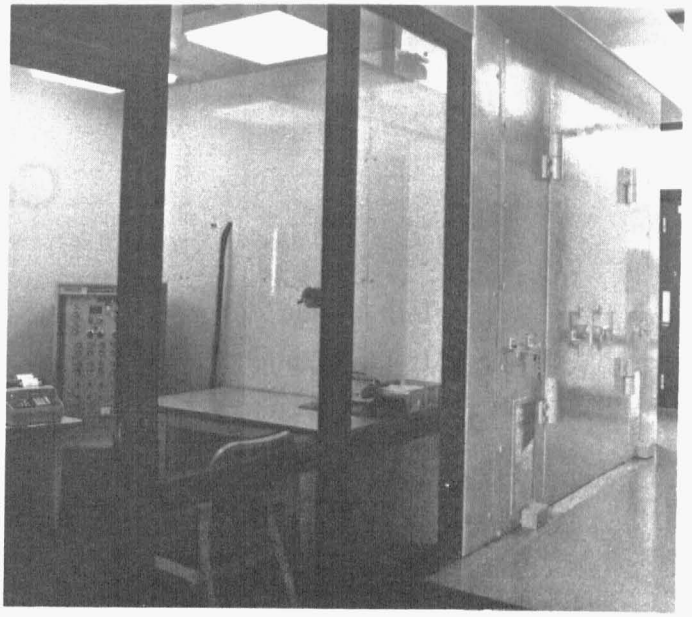
Manual for Use of

Two-Pi Liquid Scintillation Whole Body Counter

by William J. Coffman



Counting room, left, and entrance to counting chamber.

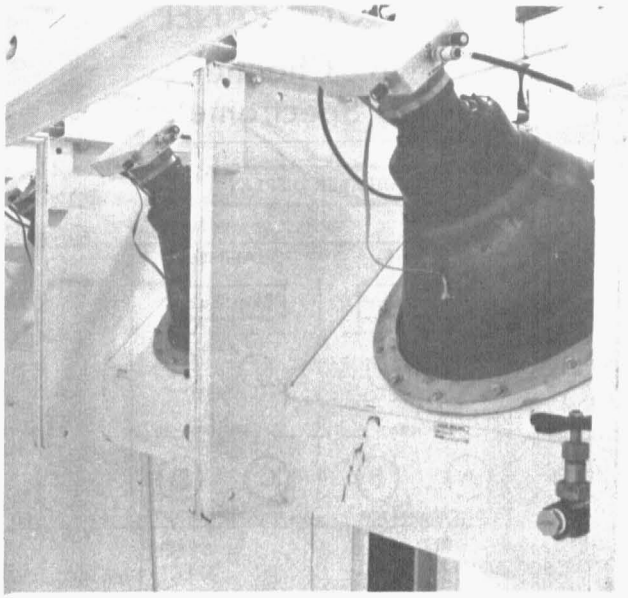
The whole body counter is a modular 2-pi liquid scintillation detector. It is housed in a steel room. The six detector tanks are constructed of 16-guage (1.52 mm or 0.0598 in.) stainless steel. On the back of each detector tank is a 40.6 cm (16 in.) DuMont-K-2128 photomultiplier tube (PMT) and a pre-amplifier.

The six tanks are arranged in pairs to form a half-cylinder 2 meters (78 in.) in length and 61 cm (24 in.) in diameter. Each tank is 66 cm (26 in.) in length. Approximately 87 liters (23 gallons) of scintillation liquid composed of a toluene equivalent solvent, PPO, and dimethyl POPOP fill each tank.* The tanks are coated inside and outside with Packard White Epoxy Paint.

The detector can be moved in a vertical direction, and each tank can be moved horizontally. The detector is shielded by an 80 metric-ton (88T) pre-World War II steel chamber whose walls are 10 cm (4 in.) thick, and whose floor and ceiling are 15 cm (6 in.) thick. The dimensions of the steel room are 2.7 meters high (9 feet), 2.7 meters wide and 5.5 meters deep (18 ft.). Additional shielding is provided by 40 metric-tons (44T) of high purity quartz sand placed over the top of the room and along the north and south walls. The sand acts as an attenuator for cosmic radiation, and has been effective in reducing the background radiation by about 17 percent.

Contamination from soil potassium, radium, and fallout is a constant problem. Visitors and researchers are requested to remove their shoes when entering the building by the south entrance. Also, domestic animals should always be washed with a detergent before beginning a whole body count. People who are to be counted are asked to remove street clothing and wear one of the paper gowns which are provided. Appendix A includes a list of rules and explanations used to help researchers formulate their experiments. (See section on calibration and counting procedure.)

* PPO is diphenyloxazole and dimethyl POPOP is 1, 4-bis-2-(4methyl-5-phenyloxazolyl)-benzene.



Backs of three tanks showing PMT tubes and preamplifiers.

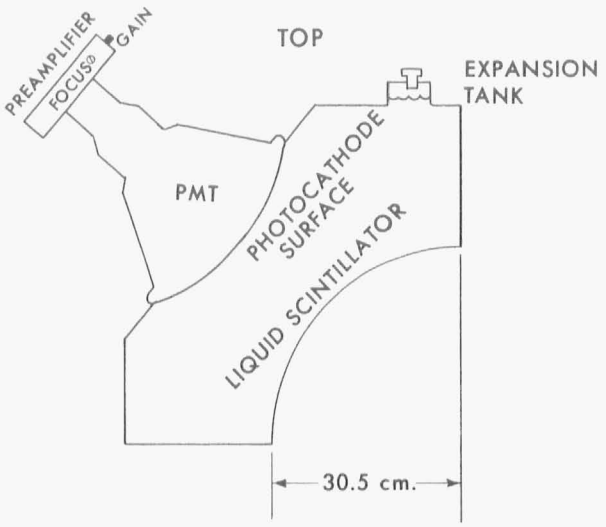


FIGURE 1 DETECTOR TANK PHOTOMULTIPLIER, AND PREAMPLIFIER

Instrumentation

The photomultiplier tube (PMT) is a sophisticated photoelectric cell. A gamma ray, i.e., photon, passing through the liquid scintillator, loses energy mainly by Compton scatter. Compton scatter is characterized by a photon (energy range is approximately 20 kev to 30 mev) colliding with an atom or molecule of the liquid and the subsequent transfer of some of its energy to an orbital electron. Scattered electrons are called Compton electrons. The gamma ray is reduced in energy and scattered in the liquid to collide with other atoms. A full transfer of energy can take place only by multiple collisions with the atoms of the scintillator.

The Compton electrons lose their energy by producing light photons or scintillations in the liquid. The light photons eventually reach the surface of the PMT and initiate a photoelectron from the surface of the photocathode. The photoelectron is multiplied through several electronic stages within the tube and eventually emerges as a volley of electrons. The electrons pass through a pre-amplifier which amplifies the electronic signal before it reaches the recording equipment (Figure 1).

The energy peak in a liquid system will be different from the energy peak produced by a crystal detector. The difference is due to the mechanism by which the gamma ray loses its energy in the two different detectors.

The energy peak from Compton electrons in liquid scintillation can be determined by the formula

$$E(\text{mev})^{**} = \frac{E_1}{1 + \frac{0.51}{2E_1}}$$

where E is the maximum energy of a single Compton scatter produced by a gamma ray whose peak energy is E_1. Accordingly, the energy peak for ^{137}Cs would be 0.480 mev rather than 0.662 mev. Potassium-40 would be 1.24 mev rather than 1.46 mev. (Fig. 4).

** Mev = Million electron volts, a unit of energy.

Spectrometer

The recording equipment consists of a two-channel, gamma spectrometer (Figure 2). The *Background Subtract* units (*Bkg. Sub.* in Figure 2) on the spectrometers are not used with the whole body counter. Likewise, the *Reject* should be left in the OFF position since this control knob is used with the *Background Subtract.* The *Minute-Second* (*Min.-Sec.* in Figure 2) selector should point to 0.01 if time is in minutes and to 0.1 if time is in seconds.

To check the scaler portion of the spectrometer, a 60 cycle test is provided. The scaler can be checked by turning the *Mode Selector* to "Check" and *Preset Time* to 1. Thus, in one minute, 3600±6 counts (cpm) will appear in the number display.

During the 60 cycle test, do not manipulate any of the switches. Movement of the switches may present a number greater than 3600±6 cpm. For normal operation the *Mode Selector* should be set between "Auto" and "Check" on "Manual."

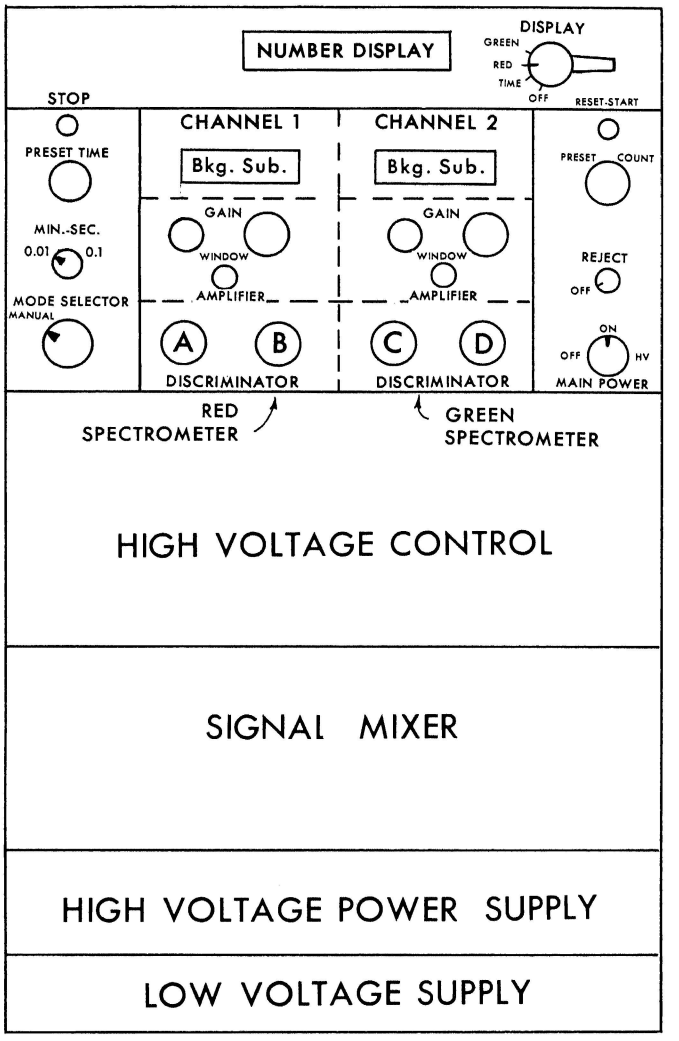
The scaler can be operated *Preset Time* or *Preset Count.* Whichever mode is selected, the one not used should be set at its maximum setting. Thus, if *Preset Time* is used, *Preset Count* should be set at 900,000; or if *Preset Count* is used, *Preset Time* should be set at 10^4. Both *Preset Time* and *Preset Count* are operational during counting. Whichever is reached first will terminate the count at that setting.

Counts accumulated in either the red channel or green channel can be read by turning the selector switch *Display* to green (..) or red (.). While performing the count, care should be exercised when moving this switch; noise, i.e. counts, can be introduced into the gross reading. The *Display* mode allows the operator to observe the elapsed counting time.

The *Amplifier Gain* switches for the *Red* and *Green Spectrometers,* have a coarse and fine adjustment. The coarse adjustment center can be moved to either "10%" or "100%." If it is on "10%," each division is 1 and any number up to a maximum of 10 can be selected. The fine adjustment allows selection from 0.1 to 1. If the coarse is set on "100%," each division is 10 up to a maximum of 100, and the fine adjustment is from 1 to 10.

After the spectrometer has been standardized (see section on Balancing and Standardization), whatever the *Gain* setting for the red channel, the *Gain* setting of the green channel will be half this

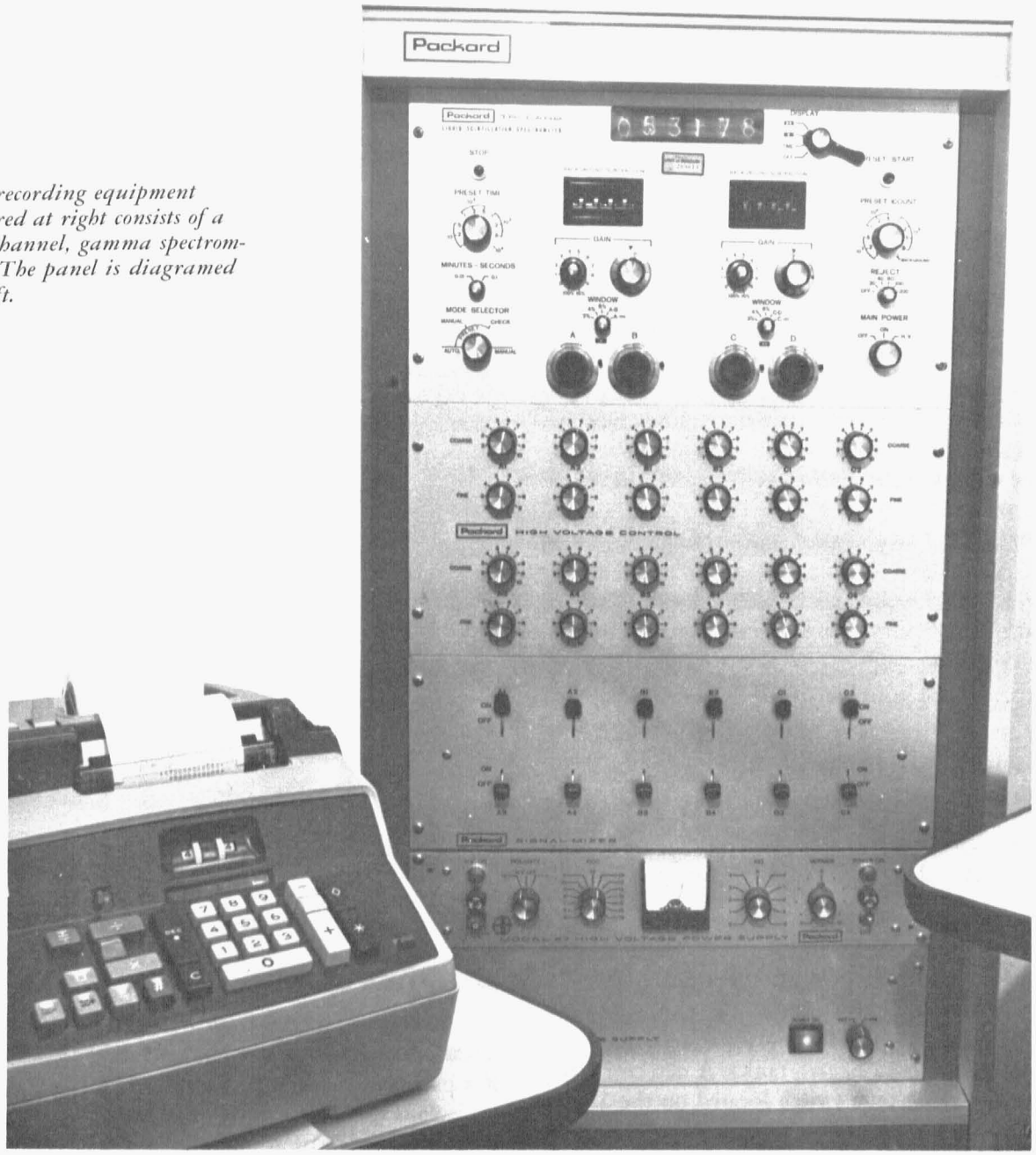
FIGURE 2
CONTROL PANEL
Two Channel
Gamma Spectrometer



value. For example, if the *Gain* in the red channel is 26, the *Gain* in the green channel will be 13.

The full scale limit of the *Discriminator* in the green channel (D) will be twice that of the red channel (B). For example, if the scale unit is in energy, and the "B" pot is equivalent to 1.0 mev when turned clockwise to its maximum position (10.0), then the full scale in the green channel will be 2.0 mev at 10.0.

The width of the discriminator window is con-



The recording equipment pictured at right consists of a two-channel, gamma spectrometer. The panel is diagramed at left.

trolled by the dial indicated as *Window* immediately above and between the two pots in the red and green spectrometers. The *Window* control has settings at "2%," "4%," "8%," "A→B" ("C→D"), and "A→ ∞ " ("C→ ∞ "). For normal operation the settings are "A→B" or "C→D." At these settings, only events occurring between the limits set at "A" and "B" or "C" and "D" will appear in the number display. When the knob is turned to "8%," the window is set from "A" or "C" to 8 percent of the maximum value of whatever the calibrated scale happens to be. If this was 1.0 mev (1000 kev†) in the red channel, the electronic window would be set to read from "A"→"A" + 80 kev. Or, if the reading was in the "green" channel, and the maximum energy was 2.0 mev (2000 kev), the electronic window would be set to read from "C"→"C" + 160 kev. If it was on "2%" and on "red," the window limits would be

†Kev = Kilo electron volts, a subunit of the mev. One thousand electron volts.

"A"→"A" + 20 kev, etc. The "B" or "D" pot should be set far enough ahead of "A" or "C" pots so that they will not cut off the effect of the "8%" window.

If ^{137}Cs standard is placed under the detector and the *Window* is reduced to "4%," the highest count rate will occur when "A" is at 4.60, provided the "red" channel is standardized to a maximum scale of 1.0 mev. If the *Window* is "8%," the highest count rate will be set at 4.40. Remember that the energy peak for ^{137}Cs is reduced to 0.48 mev. Therefore, the window limits are set for 4.60→5.00 and 4.40→5.20 for "4%" and "8%" *Window* settings.

In the green channel the limits for a "4%" and an "8%" *Window* would be 2.00→2.80 and 1.60→3.20, respectively. Recall that the amplifier gain for the green channel is half of that of the red channel, but the energy scale of the green is twice the value of the red.

Whenever any of the control knobs in either the discriminator *Gain* or *Window* are moved, the operator should wait three minutes before beginning a count. The validity of the counts prior to this electronic equilibrating period will be questionable.

Electronic windows can be set so that lower and upper spectral limits can be established for different radioisotopes. Only counts occurring within these set limits are recorded. The window limits for ^{131}I are 1.3 at "A" and 7.0 at "B" in the red channel. The window limits for ^{40}K are 5.0 at "C" and 10.0 at "D" in the green channel. The energy bands being recorded are 0.13 to 0.7 mev and 1.0 to 2.0 mev, respectively. All other energies in the two spectrometers will be discriminated against. Other window settings in Appendix B.

The detector is divided into three sections. Each section is composed of two tanks. A detector tank can be turned *On* by a switch located on the *Signal Mixer.* When the switch is up, the scaler of the spectrometer will receive a signal from the designated tank. There are 12 switches on the *Signal Mixer* panel. Only those switches on the upper half of the panel are operational. The bottom half is not connected. The switches are labeled A-1, A-2, B-1, B-2, C-1, and C-2.

The A tanks are located next to the east door of the chamber. The B's are the middle tanks, and the C's are the tanks on the west end (old livestock pavilion side) of the room. Switches A-1, B-1, and C-1 control the three tanks on the south side (recording room side) of the chamber, and switches A-2, B-2, and C-2 control the tanks on the north side. This arrangement constitutes the modulation of the detector.

The basic high voltage to the PMT is controlled by the *High Voltage Power Supply.* The high voltage has been set at 1300 volts. The operator should check this setting occasionally and be certain that the *Polarity* control knob is on positive (+) only.

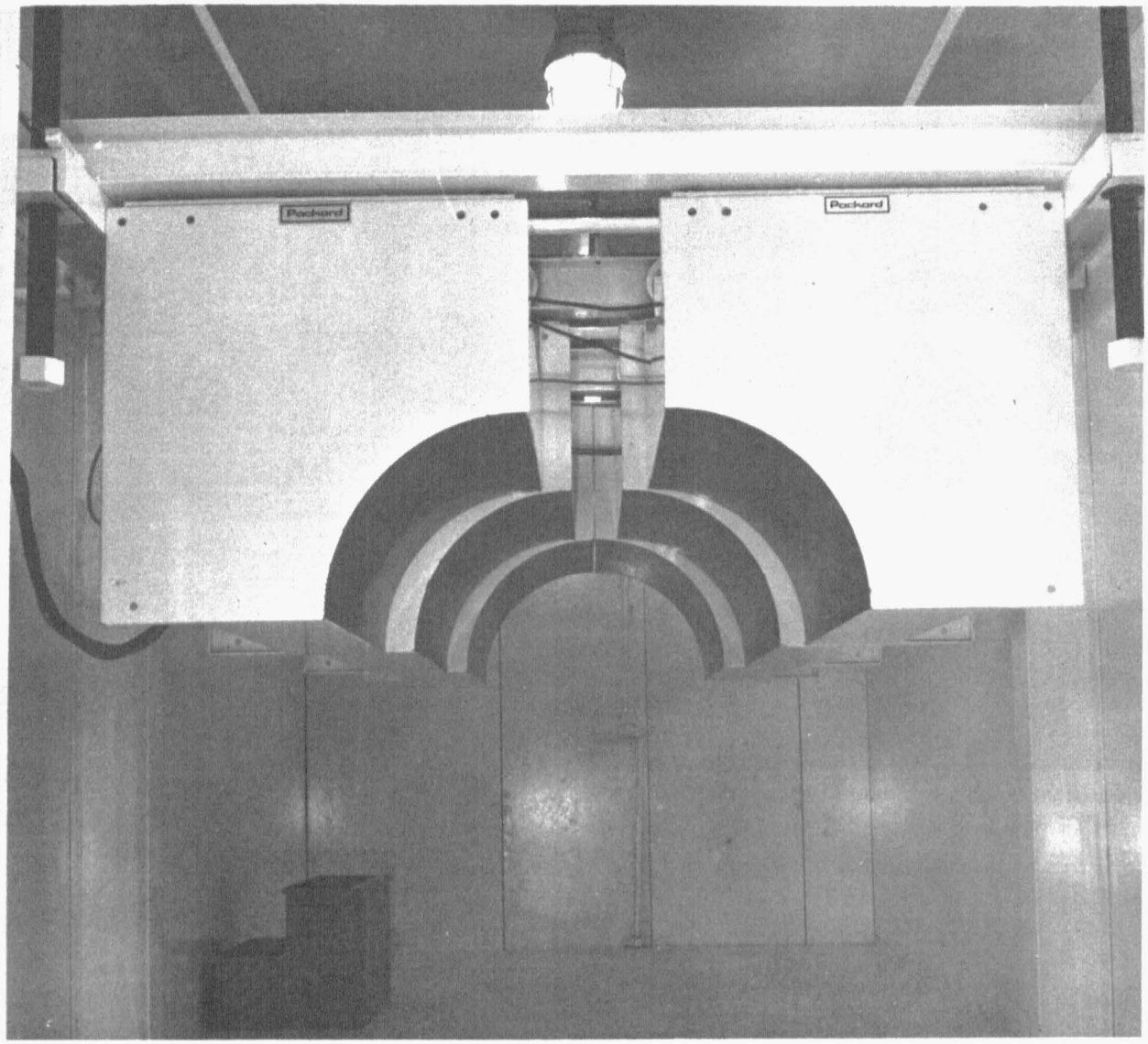
Regulation of the high voltage is by coarse control (*X100*), fine control (*X10*) and very fine control (*Vernier*). The coarse control advances through steps of 100 volts to a maximum of 1900. The fine control advances through steps of 100. The *Vernier* is graduated in units of 1 volt to a maximum of 10. The maximum capable voltage capability would be 2110. It is not likely that high voltage requirements will be greater than 1500 volts. If the power should be shut off or go off for any reason, such as power shortage, changing of cables on the instrument, or replacing parts, turn the coarse control back to zero. This will prevent damage to instrument parts when the power is turned back on (prevention of power surge). After the power is ON, advance the coarse control back to its original setting. For present conditions, this would be 1300.

High voltage control to each PMT is regulated at the *High Voltage Control* panel. Although the basic high voltage is 1300, each PMT has a different high voltage and must be standardized so that when all six tanks are operated together, all PMT's will be balanced and will function as one detector.

At the rear of the *High Voltage Control* case, the operator can, if he wishes, transfer cable leads. Thus, if a particular tank is performing badly, an attempt to isolate the problem by running the cable lead through a different set of high voltage control potentiometers can be done.

For example, during the early periods of checking the counter out, it was learned that C-1 performance was questionable so it was transferred to C-3. High voltage control for C-1 is (1/68) controlled through C-3.

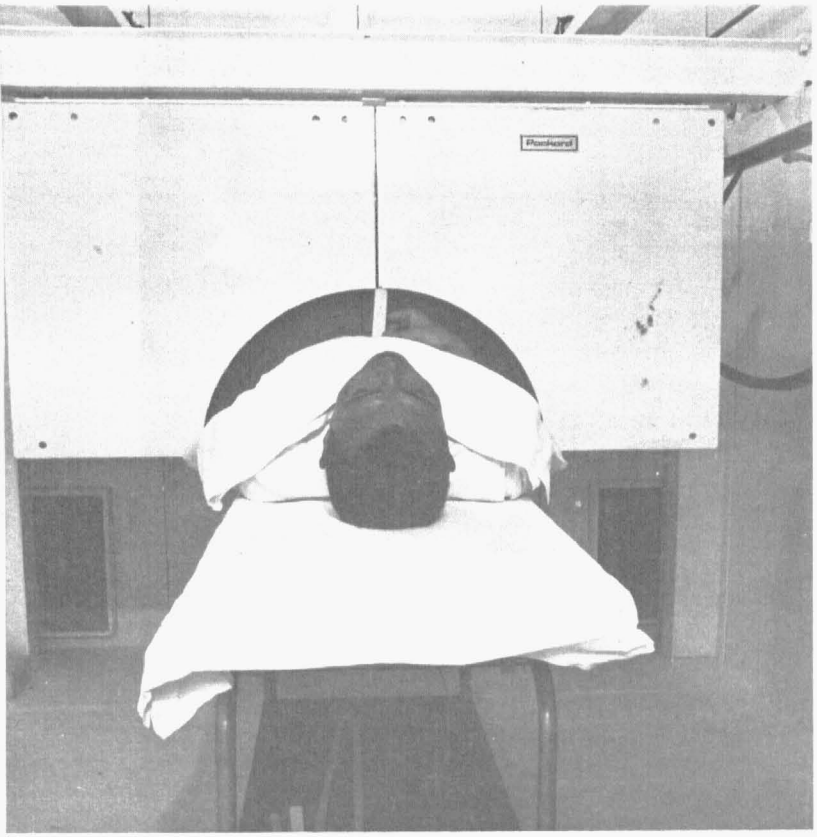
Substitution can be very helpful for solving problem areas. However, the end pots (A-1 and C-2)

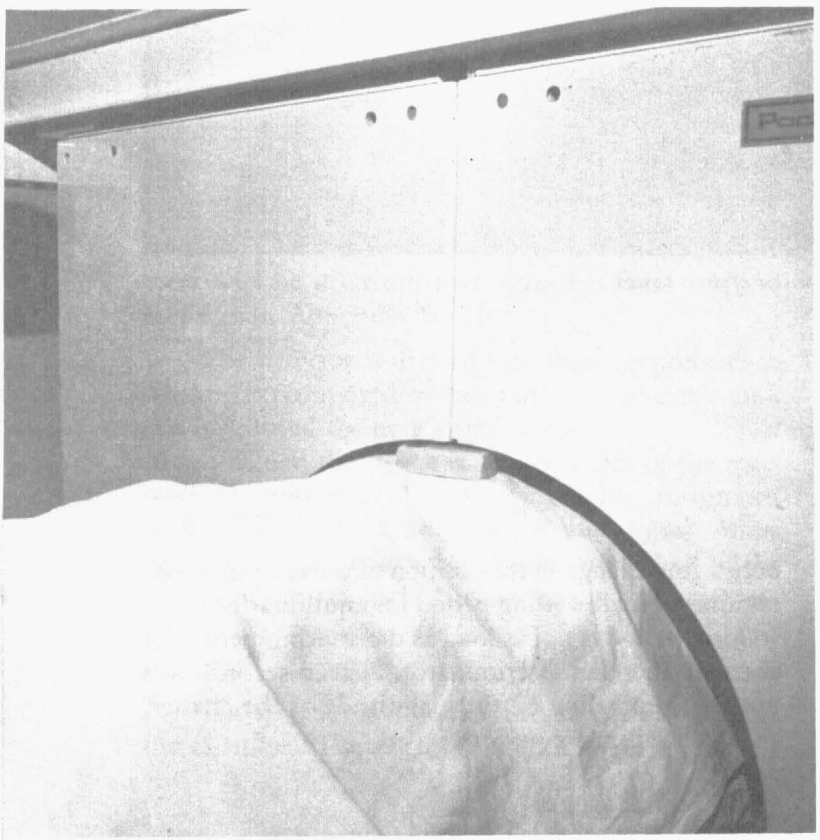


The detector is divided into three sections. Each section is composed of two tanks.

cannot be transferred electronically to the lower part of the control panel (A-3 and C-4).

The high voltage control for each tank is checked at weekly intervals and a record is kept. Large changes in high voltage requirements may be indicative of serious problems. Minor changes are expected and will occur. Any change in the position of the control knobs requires a small waiting period for equilibration. This waiting period is not as long as the waiting period for the amplifier and discriminator. Fifteen seconds to a minute, depending on the magnitude of the change, will suffice in most instances.





Human adults are counted anterior-posterior with a 10.16 cm (4 in.) DSD. A child's position (below) for ^{40}K is posterior-anterior and the DSD is measured from the buttocks at 2.54 cm.

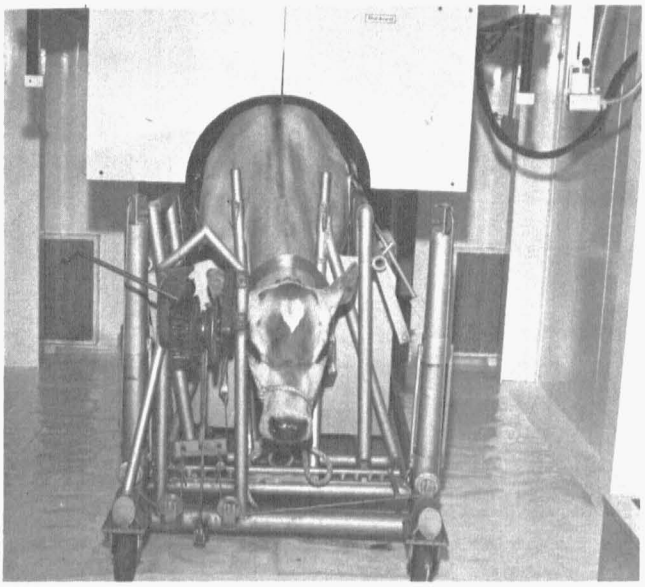
Geometrical Considerations

One of the advantages of a modulated detector is that it will permit compartmenting. This allows the operator to adjust the sensitivity to suit the sample size. Little would be gained by counting a rat with all six tanks and suffering the consequence of a high and variagle background. For this purpose the best overall arrangement can be achieved by using two tanks.

A subject must be at least 132 cm (52 in.) in length for full utilization of all six tanks. Since each of the three sections is 66 cm (26 in.) in length, a subject larger than 132 cm would extend far enough to require all detectors. For objects less than 132 cm, it is better to use four tanks, i.e. only two sections.

Optimal geometry is obtained by placing the subject to be counted in the center and as close as possible to the top of the detector. The center of the detector is approximated; however, the measure called the top of the ***detector to the subject distance*** (DSD) is critical. One must realize that the objects being counted are irregular in shape and size. An error of ± 2 mm is tolerable because of this irregularity. Human adults are counted anterior-posterior with a 10.16 cm (4 in.) DSD. This may not meet the optimal conditions, but it is more comfortable and presents fewer problems psychologically to the subject.

A 10.16 cm DSD has been used for animals. Again, this may not represent the best, but it allows for sudden movements of the animal. An excited animal could rupture a tank if it were close enough to it.



Cow in position for counting.

The 10.16 cm for humans greater than 132 cm is that distance from the sternum (breastbone) to the top of the detector arch. For animals it is the highest point on their backs to the top.

Children will have the same DSD as adults except when determining ^{40}K. A child's position for ^{40}K is posterior-anterior, and the DSD is 2.5 cm, measured from the buttocks.

Detector efficiency is reduced as the activity moves out and away from the detector. A person can be in the steel chamber and not significantly influence the count rate of the subject if that person is is at least 7 feet from the end of the detector. Ideally, the person should be at the other end of the room, but children and some animals respond to their environment better if they can see or hear someone in the room with them.

Balancing and Standardization

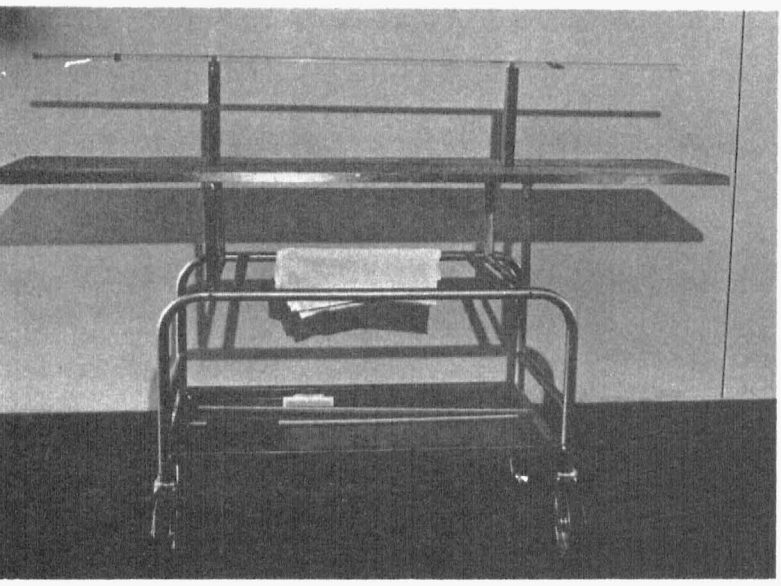
Balancing: Balancing requires a certain amount of skill that can only be acquired by experience. It must be remembered that the original signal from the photomultiplier tube is very weak. This signal must be amplified, or in other words, it must "gain" amplitude to be recorded.

There are three gain control adjustments:

1. The gain control of the *pre-amplifier* (Fig. 1).
2. The gain control of the amplifier of the *spectrometer* (Fig. 2).
3. The "Coarse" and "Fine" adjustments on the *High Voltage Control* panel.

The preamp has two variable dials accessible to the operator. One is the *Focus* which maximizes the flow of electrons from the cathode of the PMT. The other dial is the *Gain* which serves as a proportionality variable to maximize the signal (net counts) to noise (background counts) ratio, S/N. Both dials are at their optimum when they are set at 50% of rotation. This is true for any one tank, but may not be true when balancing all six tanks.

The integration of all six tanks to function as one large detector may require a loss of gain in several of the preamps. Unlike the perfectly matched PMT in a Packard Tricarb, the PMT's of the whole body counter are not matched. They are the largest PMT's made, and are made only upon special order. Matching of PMT's demands a large production of tubes.



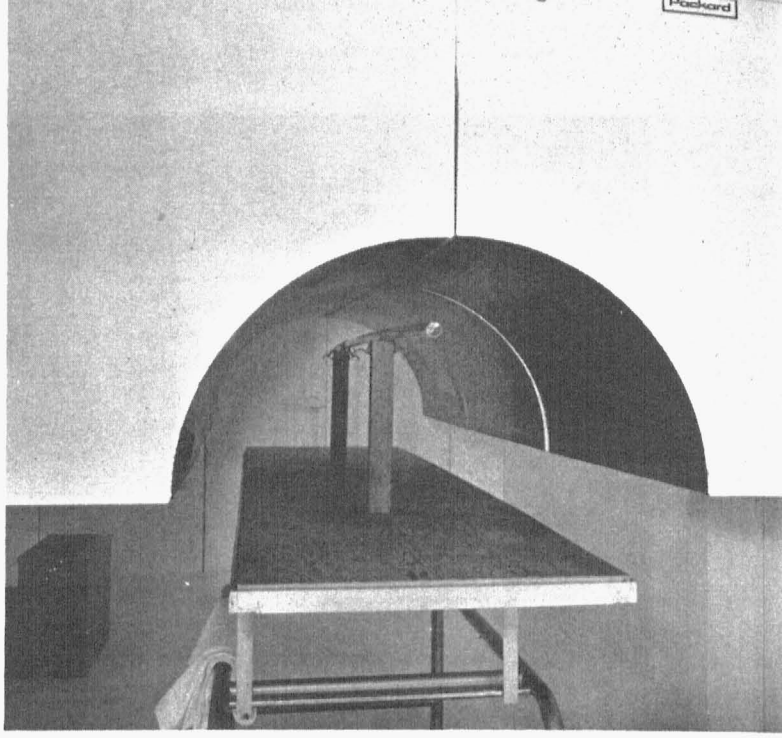
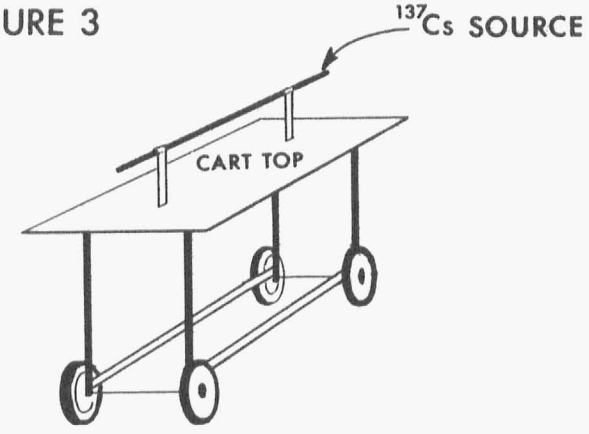


FIGURE 3

The standard is held in place by two upright supports on top of a mobile cart.



μC_i*. The activity is as high as practicable in order to receive high count rates in a narrow spectrometer window for short counting times.

The standard is held in place by two upright supports on top of a mobile cart (Fig. 3). When the detector is raised to the height indicated by the mark at the east door as "^{137}Cs rod," the standard is at its optimal distance from the top of the detector.

To keep consumer costs down, only those necessary for the operation of the system are made. (How many institutions have whole body counters utilizing a 40.5 cm PMT?) Therefore, the *Focus* and *Gain* of the preamp are used to help match or balance the PMT. The 50% position is the starting point, but, in actual practice, the dials may end up at some other location.

The standard is a ^{137}Cs liquid source in a 1.83 meter (6 ft.) plexiglass tube. Cesium-137 is a frequently used standard because of its physical half-life (30 yrs.), and its monochromatic gamma ray energy (0.662 mev). The activity in this standard is about 2

* μC_i = Microcurie which is 1/1,000,000 of a Curie. The Curie is 3.7×10^{10} disintegrations per second.

Procedure

(See photo, page 7)

1. Turn all of the "Coarse" and "Fine" control knobs on the *High Voltage Control* panel to the same dial setting, of coarse 10, fine 10.
2. Select any one of the 6 tanks and turn this unit *ON* at the *Signal Mixer.*
3. The *Gain* Control of the "red" channel should be at "100%," and the *Window* should be at "8%." Set *Preset Time* to "0.1." Set the *Gain* in the "green" channel to "100%" and the *Window* to "2%." The coarse and fine adjustments of the *Gain* control should be set at zero in both channels.
4. The ^{137}Cs energy peak will appear at 0.480 mev. This will be 4.80 on the "A" discriminator after standardization. Therefore, for an "8%" window, the highest count rate would be at 0.440 mev or 4.40 on the "A" discriminator (4.40 - 5.20). This is true when looking at the peak for all six units, but when working out of any one unit, the peak appears at a dial setting of 4.20 on the "A" discriminator. The reason for the difference between dial settings when using six units and one unit is not known. At this stage of balancing, the peak is not a critical setting, as will be learned later on. Therefore, set the "A" discriminator at 4.80. The "B" discriminator should be opened to 10.0.
5. Adjustments of the *Focus* and *Gain* of the preamp may be necessary. Leave the steel doors open during balancing to allow frequent and easy ac-

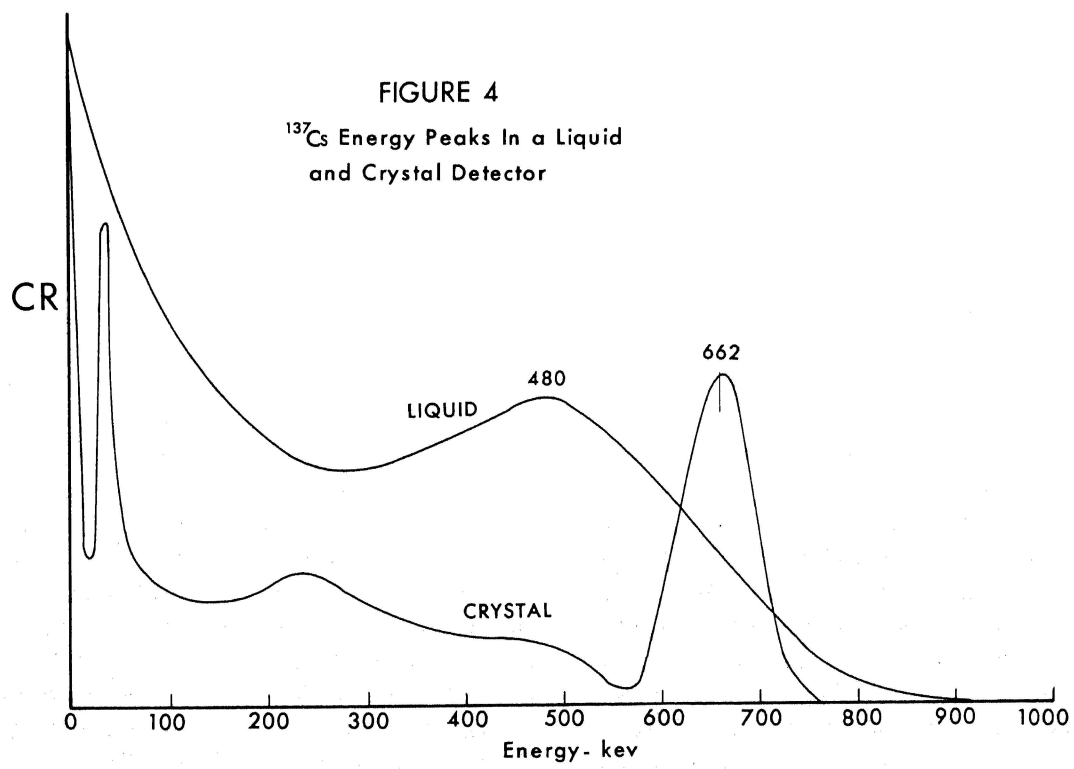


FIGURE 4

^{137}Cs Energy Peaks In a Liquid and Crystal Detector

cess to these controls. Only the gross count rate (no background correction) is necessary for the balancing procedure. With the ^{137}Cs standard in place, advance the coarse control of the amplifier gain, taking counts for 0.1 minutes at each advancement. The operator will observe that at one particular setting, the count rate will be higher than for the other divisions of the coarse gain. This is the peak. If a peak is not obtainable, go to the preamp and turn the preamp *Gain* counter clockwise in very small increments until the count rate passes through a peak at the amplifier *Gain.* Preamplifier adjustments are not necessarily the rule. If required, the task can become tiresome.

After passing through the peak, set the coarse gain of the amplifier at that setting where the peak was observed to fall, and advance the fine gain to "zero in" on the peak. The counting period may need to be advanced from 0.1 minute to 0.5 or 1.0 minute to observe noticeable changes in the count rates for the various fine control settings. It is usually desirable to advance the fine control dial through increments of 10, then about 4 readings at increments of 5, and finally about 10 readings at increments of 2.

6. *Do Not Change the Amplifier Gain.* Determine what value discriminator "A" will have for the other 5 units. That is, determine the energy peak for the other 5 detectors through discriminator "A." Record these values. All units should be OFF at the *Signal Mixer* except the one for which the peak is being sought. Place discriminator "A" at the lowest of the six recorded discriminator values.

7. Movement of the "coarse" and "fine" controls on the *High Voltage Control* panel clockwise will shift the energy peak to the right. Likewise, rotation counterclockwise will shift the peak to the left. Since the "coarse" and "fine" control dials are already at their maximum, a peak appearing at a lower value cannot be brought up to match one which has a higher level. Therefore, select the detector with the lowest value and adjust the "coarse" and "fine" control knobs of the other five detectors counterclockwise to match the peak at the lower setting. It is not likely that the detector which was used to determine the amplifier gain will be the one with the peak at the lowest value.

   In this last step the amplifier gain remains unchanged, and discriminator "A" is set at that setting where the lowest peak occurred. It is not necessary to be exact at this point.

8. To calibrate the discriminators to an energy scale of 0 to 1.0 mev, move discriminator "A" to 4.40.

9. Through the fine adjustment of the amplifier, and with all six detectors ON, peak the system to 4.4.

10. At the *Signal Mixer,* turn all of the detectors OFF. Turn A-1 ON. Set *Preset Time* to 0.5. Record half minute readings at increments of 0.10 from discriminator "A." When the individual detector is calibrated in energy units of 0 to 1.0 mev, the peak will set at 4.20 $\pm$ 0.1.

    Repeat this last step for each detector.

11. With *all* detectors on, *Preset Time* 0.5 and "8%" *Window,* the energy peak should occur at 4.40 $\pm$ 0.1. With practice, the operator can take three readings, one each at 4.2, 4.4, and 4.6 and determine the peak. The counter is now as near perfectly balanced as possible.

Steps 10 and 11 of the standardization procedure should be repeated at the first of each week, usually on Monday. A record of the *High Voltage Control* settings for each tank and photomultiplier tube should be kept. Once the amplifier gain has been set, it should not be changed. The amplifier and *High Voltage Control* can be regulated by the *Gain* of the preamp. Extremely high noise counts can be reduced by decreasing the *Focus* of the preamp. It is not recommended, however, that the *Gain* or *Focus* of the preamp be changed, since any changes will result in changes at the control panel. If at all possible, avoid any changes at the preamp!

After standardization, the energy scale can be changed by changing the amplifier gain at the control panel. If the scale is 0 to 1.0 mev in the red channel, it can be changed to 0 to 0.5 or 0 to 2.0 mev by either halving or doubling the amplifier gain setting. Remember that whatever the gain is in the "red" channel, the *gain* in the "green" channel is half that of the "red" and the *energy scale* is doubled.

Calibration and Counting Procedures

To calibrate the counter for a particular radioisotope, consideration should be directed to the quantity of the isotope needed for measurement, major gamma rays involved, the spectral shape of the isotope,and the size and shape of the object or subject to be counted.

An advantage of liquid scintillation detectors is their ability to detect small amounts of radioactivity in short counting periods and with low statistical errors. In addition, their counting efficiency is much higher than that of other types of detectors. These are very important qualifications when measuring an isotope such as ^{40}K, whose concentration is minimal to begin with.

A disadvantage of liquid scintillation detectors is their inability to resolve energy peaks. Figure 4 shows the energy peaks for ^{137}Cs for a crystal detector and the University of Missouri's liquid counter. The curve demonstrates that because of the poor peak associated with the liquid counter, an alternate method for determining the resolution must be used.

Typically, the resolution for a detector is defined as the *full* energy width of the peak as measured at *half* the maximum count rate divided by the peak energy, and expressed as a percent. The width of the energy peak for the liquid detector is too broad. The resolution for the liquid scintillation is, therefore, re-defined as *half* the energy width of the peak measured at half the maximum count rate, divided by the peak energy, and expressed as a percent.

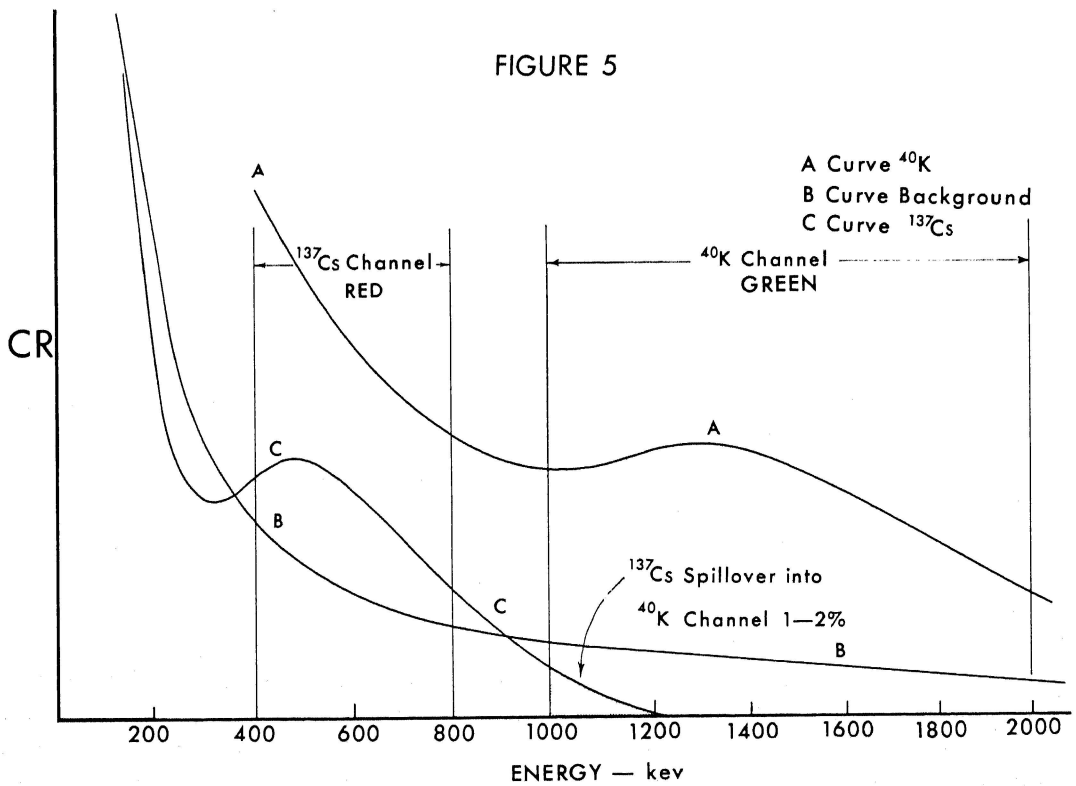
The established practice for determining the optimal window setting is to select the highest figure of merit (FM) or signal to noise ratio (S/N). The FM is expressed as the ratio of the counting efficiency squared to the background.

$$FM = E^2/Bkg.$$

The counting efficiency E is the ratio of the observed net count rate to the disintegration rate. The S/N is the ratio of the net count rate to the background count rate.

Either method for window selection is acceptable. However, because of the poor resolution of the liquid counter, and because of the high PMT noise level at the lower end of the energy scale (Figure 5), these methods may not be the *best.* For example, the

FIGURE 5



FM study for ^{40}K suggests that the optimal window is 3.30 to 10.0 in the green channel. However, ^{137}Cs persists as a contamination problem, and with a window of this width, considerable spillover into the ^{40}K channel occurs. If the window width is reduced to 5.0 - 10.0, spillover of ^{137}Cs into the ^{40}K channel is insignificant and can be ignored. This narrow window reduces E, but a small reduction in E is a better choice than trying to solve a complex equation for spillover. For younger subjects, where ^{137}Cs has not accumulated to any significant degree, the wider window is preferable when determining ^{40}K.

Animal and sample cleanliness is very important. Samples free from outside contamination (fallout, ^{40}K from soil, radioactive materials from other experiments, etc.) are desirable for consistent results. This is particularly true for ^{40}K and ^{131}I determinations.

Potassium-40 values will be influenced by dirt that is brought in with the animal from the yard. Animals should be washed with a low potassium detergent and rinsed at least twice with water before counting. If a detergent low in potassium is not available, several rinses following the wash may be satisfactory.

As Lohman *et al.* have pointed out, the gastrointestinal fill of ruminants does influence the ^{40}K count rate (3). Since feeds are usually quite high in potassium, it is suggested that animals be placed on feeds low in potassium four or five days prior to performing a potassium analysis.

Experiments using ^{131}I need careful planning, since any loss of radioactive iodine in the counter, on the animal, or on the animal holder will give erroneous results. This is true of all experiments where the radioactivity may contaminate areas where it is not wanted. However, the physical and chemical properties of iodine make it more difficult to clean up, and any small amount of the element released could be detectable. Rigid controls are necessary to prevent contamination.

The laboratory is not equipped to assay materials before they are used. All too frequently the suppliers' assay values will be slightly off, either higher or lower than reported, or the investigator may have made an error in calculations or dilution. A check should be run on the material before it is administered. The check is made by placing about 0.5 microcurie (μC_i) of the intended liquid or dissolved material in a 1 or 2 liter container, bringing it up to volume with water and counting in the whole body counter. Except in cases where very short half-lives are involved, maximum activity levels for animal experiments should be kept between 1 and 2 μC_i. The maximum number of counts that can be recorded by the scaler is 900,000. Counting times less than 1 minute that give this level of count rate adversely affect the stability of the photomultiplier tubes.

Karzmark has reported on photomultiplier tube fatigue effects (2). Photomultiplier fatigue and recovery effects, i.e. gain variations, are complicated and different for various photomultiplier types and can differ from tube to tube within a group. Tubes may require considerable time for recovery after the exposure to high scintillation rates. Length of exposure may also influence the recovery period. Gain variations appear to be the result of electron bombardment of the sensitive secondary emitting surfaces, and can be greatest in the late stages where the currents have increased.

This problem is circumvented by restricting the count rate to the maximum counting capacity of the scaler for a counting time of greater than one minute. Potassium-40 levels in vivo present no problems, but when performing animal experiments where radioactivity may be introduced, a maximum level of activity should be specified.

Counting rates can be reported in any set of values the researcher desires. For high counting rates, counts per second are more convenient since the values will have fewer significant figures. Conversely, for low counting rates, counts per minute are preferable.

The scaler has the capacity for recording events as *preset count* or for a *preset time.* When making a series of measurements, a comparable precision of each measurement is usually desirable. By presetting a number of counts, the precision of the observations is also preset. By measuring the time required to accumulate a preset number of counts, a constant counting error can be maintained. Error in the measurement of time for the preset count method cannot be neglected when time intervals become short. Thus, the preset count setting should be selected in such a way that this error becomes small. As a rule, when time intervals of one minute or longer are selected, the error need not be accounted for. It has been observed that the *preset count* method is limited by the accuracy of the time measurement. It is therefore not

recommended unless methods are provided for determining time beyond two decimal places.

Sample containers, animals, etc., should have a pre-service count rate established for them. The background count rate of an object is made up of counts in the steel room R_R and counts from the object, R_o. The total background, R_B, is

$$R_B = R_R + R_o.$$

R_R will vary throughout the day, but R_o should remain fairly constant, i.e. $R_o = k$. If R_o is known prior to measuring, then during the course of the experiment, R_B can be determined by simply measuring R_R and adding k,

$$R_B = R_R + k.$$

Containers used for other experiments where radioactive materials have been employed must be screened before being used. The pre-service check will suffice for screening.

A good rule to follow when making organ comparisons is to keep sample containers equal. For example, if ^{59}Fe is used in an animal experiment, and it is of interest to know how much ^{59}Fe will be found in the liver after a certain mixing time, obtain two containers equal in size. In one place a dose of ^{59}Fe equal in activity to that which was given originally. Weigh the liver, homogenize it and re-weigh the material. Fill the second container with the mixed liver, and count both containers. Containers should be plastic and not capable of leaking when on their sides. Results can be expressed as *activity* per unit weight of sample or *count rate* per unit weight of sample. Variations of this procedure can be applied to the situation that best fits the experiment.

^{40}K Calibration

Calibration curves for ^{40}K are constructed by counting a known amount of KCl and determining the efficiency (E) for various sizes of mock subjects. The mock subjects, which are known as phantoms, can be fashioned from various materials. Some people mix the KCl with sugar and load it into sacks that have a negligible background or they may use cardboard boxes. The containers should be small enough so that different sizes and shapes can be formed to represent the subject being studied.

Another method is to mix the KCl with water. This is the method used in this laboratory. The loading of the various water phantoms and their absolute count rates are listed in the appendix (Appendix C). Calculations for determining the absolute count rate and some ^{40}K constants are also presented.

Counting efficiency may be expressed in one of two ways. In the narrow sense of the word, E is the ratio of the observed counts to the total reaching the detector. In a wider sense of the term, E is the ratio of the observed to the number of disintegrations within the radioactive sample. Since the whole body counter is a gamma counter, E will be expressed according to the first definition, because only the gamma rays are being detected.

Another ^{40}K calibration method involves mixing KCl and water. These are ^{40}K water phantoms.

Calibration curves for ^{40}K water phantoms show a plot of E vs. body weight in kgm. The efficiency is determined by taking a room background, counting a phantom three times to determine the average, then taking another room background and averaging it with the first. This average is subtracted from the average gross count rate of the phantom. The section on counting statistics will explain why this procedure is followed.

If no major changes occur in preamp setting (*Gain or Focus* change can be considered a major

change unless proven otherwise), the calibration curves for the different geometry conditions are not re-determined, but merely checked against a standard phantom size. When all six detectors are used, the standard phantom size is 66 kgm; for four units, the size is 21.8 kgm.

The ratio of efficiency of the standard phantom (E_2) to efficiency from the curve (E_1) provides a correction factor. If the standard phantom (E_2) deviates by more than ± 0.5% (one standard deviation) of the value from the curve (E_1), repeat the efficiency determination. If the deviation is consistent, multiply the values from the calibration curve by the correction factor E_2/E_1.

If the difference in self-absorption of the gamma rays from the two isotopes is ignored the determination of grams K by the internal standard method is as follows:

$$\text{grams K subject} = (^{40}\text{K subject cpm}) \text{ X}$$

$$\text{X} \left(\frac{\text{gms K bottle}}{\text{cpm } ^{40}\text{K bottle}} \right) \text{ X} \left(\frac{\text{cpm } ^{42}\text{K bottle}}{\text{cpm } ^{42}\text{K subject corr.}} \right)$$

Calibration curves using KCl assume that there is a direct relationship between the homogeneous potassium phantom and the heterogeneous whole body. Some investigators feel that an internal standard provides a greater confidence in the calibration and, hence, the estimation of grams K. The isotope commonly employed is ^{42}K. Potassium-42 has a physical half-life of 12.4 hours and 1.52 mev gamma, which is very close to the gamma energy of ^{40}K (1.46 mev).

The correct net cpm ^{42}K of the subject is determined according to: corrected subject cpm ^{42}K = (cpm ^{42}K subject) - (cpm ^{40}K subject). The bottles are approximately the same size and shape as the subject. The last expression is a geometrical factor which corrects for the difference in counting efficiency between ^{42}K in the bottle and ^{42}K in the subject. Correction for ^{42}K decay is pertinent.

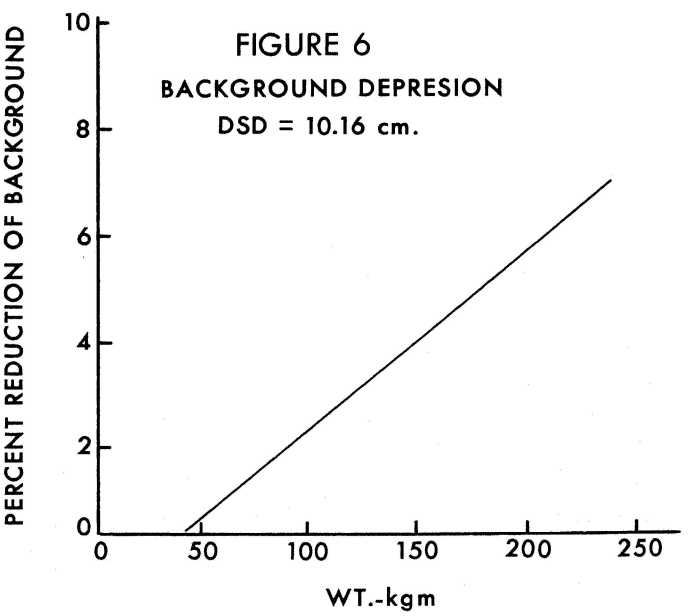
Greater accuracy is obtained by waiting for the ^{42}K to reach equilibrium in the body's potassium pool, and correcting the excretion of ^{42}K. For this procedure, two additional bottles are needed. One is used to collect urine, the other for a ^{42}K standard with which to compare urine counts. The method calls for the collection of urine 24 to 48 hours post injection of ^{42}K. The urine ^{42}K standard is 1/20 of the volume of the administered dose. It is pipetted into the standard bottle, and brought up to volume with distilled water equal to the volume of the collected urine. The percentage of the administered dose that is excreted is determined and used to correct for the amount excreted (5).

Some researchers feel that the rewards for allowing body equilibration and correction for ^{42}K excretion do not add appreciably to the calibration curve. Therefore, immediately following the intravenous injection of the ^{42}K, whole body counts are recorded.

The internal standard technique for calibrating whole body counters for potassium correlates closely with KCl phantom curves.

Electronic photomultiplier tube noise, cosmic radiation, natural radioactivity and fallout make up the background. The photomultiplier noise is next to impossible to reduce, but the background from the other sources of radiation within the existing shielding is reduced by placing a non-radioactive mass under the detector. The larger the mass, the more the reduction of the background will be. The phenomenon is due to absorption and scatter of the radiation by the object, and is known as background depression (BD).

When sample sizes exceed 200 kgm, BD should be considered since it may add to the total number of grams of potassium. Background depression is extremely difficult to determine. Many observations are needed and then one is never sure the data are consistent. *Figure 6* represents what may be BD for this detector.



To employ BD, subtract from the background the percent reduction of background in counts. This new value is in turn subtracted from the gross count rate.

$$R_c = R_s - (R_B - R_B f)$$
$$= R_s - R_B + R_B f$$

where R_c is net count rate, R_s the gross count rate, R_B the background count rate and f the percent reduction of the background for the weight of the mass under consideration.

Background depression is influenced by the size and shape of the material which occupies the space under the detector. The distance from the top of the detector to the object is also relevant.

Background depression can either be ignored or utilized in the determination of potassium. For reasons already stated, it is easier to ignore BD, but it should be kept in mind as a variable contributing to the error of measurement. When radioisotopes are administered to an animal, BD does not apply to the study, since the background of the animal is determined prior to the study and is assumed to remain constant throughout the investigation.

It has already been mentioned that contaminated street clothing, including socks, undergarments, and the fur (wool or hair) of animals, will definitely influence the estimation of potassium. Other objects which will exhibit a change in the count rate are some (but not all) of the synthetic fibers and plastics that are in the counter during the counting procedure. It is not known why some of these objects accumulate counts over a period of time, but it is reasoned that they may act like electronic scrubbers since they will develop static charges under proper conditions. In some plastics, radioactivity has been incorporated during their manufacture; it can also occur naturally. Therefore, all plastics should be checked with a pre-service count before they are used.

Porous objects, such as foam rubber or plastic foam pillows, will have variable background counts. This may be due to the accumulation of radon gas. The count rate oscillates over wide ranges when no attempt is made to account for radon build-up, between counts, in these objects. Needless to say, these materials should not be used during a ^{40}K counting procedure since count fluctuation depreciates the true value at low count rates. However, it may be necessary, for comfort, to have a hospital patient lie on a plastic foam pad during the count. Careful evaluations of the background are necessary under these conditions.

Variable ambient temperatures will be manifest by background fluctuations. This is a reason why background count rates are necessary before and after sample count rates. Ambient temperatures will vary from 21 to 24 degrees Centigrade.

If the building air conditioning should go off, counting procedures should be terminated until temperatures return to the specified range.

During the winter months, when traffic from the east entrance is likely to lower building temperatures, the door from the animal prep room to the main part of the building should remain closed. It is opened only when transporting the animal from the prep room to the counting chamber and back again. The air exchanger in the steel chamber takes air from the west side and exhausts it on the east side. Air flow is not too great, but it helps in maintaining fairly constant temperatures within the chamber. The air exchanger *should be running during all procedures.* Temperature control, like surface contamination, can never be overemphasized. Its effect is mainly on the large photomultiplier tube (PMT), and perhaps to a limited degree on the scintillator.

> As Miller & Remenchik point out, sizable errors can be introduced by small changes in the background and the gain of the PMT-amplifier system. Also, malfunction of print out system and room temperature variations will inflict errors (4). Some 16 inch diameter PMT's have a temperature coefficient of Δ 0.1% gain per Δ 1 degree F. Control of ambient temperature is obviously one control over gain stability of the PMT.

The large steel doors leading into the chamber constitute a potential source of induced variable counts. Care should be exercised to keep these doors from banging. The banging of a steel door can set up shock waves in the scintillator which could adversely affect the count rate. The animal or person in the chamber may experience some psychological discomfort which may add to the operator's problems.

Statistics

Radioactive events occur in a random manner. A radioactive atom has a 50:50 chance of disintegrating during a period of time. This period of time is known as the radioactive half-life. Fortunately, we do not deal with individual atoms, but with thousands of atoms. Statistical fluctuations change when the number of observed random events is increased. Changes are related by the "Poisson" distribution.

Symbols

- t — time.
- t_B — time to count background
- t_S — time to count sample
- N — total counts observed during some period of time
- N_B — total background counts observed during t_B
- N_S — total sample counts observed during t_S (Includes N_B)
- cpm — counts per minute
- cps — counts per second (cpm/60)
- R — count rate in cpm or cps (N/t)
- R_B — background count rate (N_B/t_B) reported in cpm or cps
- R_S — sample count rate (Includes R_B) Ns/ts reported in cpm or cps
- R_c — corrected count rate. $(R_s - R_B)$ or $(Ns/ts - N_B/t_B)$ reported in cpm or cps
- R_B^* — estimation of R_B
- R_s^* — estimation of R_s
- R_c^* — estimation of R_c $(R_s^* - R_B^*)$
- $\overline{X}$ — mean of a group of numbers
- X — a number
- n — times a consecutive number of counts were made
- σ — standard deviation $= \sqrt{N}$. If N is repeated many times, 68% of all measurements will deviate from their mean by no more than $\pm\sigma$, 95% by $\pm\sigma$, and 99.7% by $\pm 3\sigma$.
- V — coefficient of variation. Standard deviation expressed as a percent $\frac{\sigma}{N} \times 100$

  In this case N can be R_c $\overline{X}$, etc., whatever was used to determine σ
- P — acceptable percent error in a measurement reliability and confidence level.

1. Low reliability will have a confidence level of 0.68 i.e. V = P and 2 out of 3 trials will be within $\pm$ V. $N = \frac{10{,}000}{P^2}$
2. Medium reliability will have a confidence level of 0.95 i.e. $V = P/_2$ or 20 out of 21 trials will be within $\pm$ 2V. $N = \frac{40{,}000}{P^2}$
3. High reliability will have a confidence level of 0.997 i.e. V = P/3 or 370 out of 371 trials will be within $\pm$ 3V. $N = \frac{90{,}000}{P^2}$

Formula

Adding Standard Deviation from Different Variables

1. $\sigma = (\sigma_1^2 + \sigma_2^2)^{1/2}$
2. $\sigma_B = (N_B/t_B)^{1/2}$
3. $\sigma_s = (N_s/t_s)^{1/2}$
4. $\sigma_c = (\sigma_s^2 + \sigma_B^2)^{1/2}$

$$\sigma_c = (N_s/t_s^2 + N_B/t_B^2)^{1/2}$$

$$\sigma_c = \left(\frac{R_s^2}{N_s} + \frac{R_B}{N_B}\right)^{1/2}$$

$$\sigma_c = \left(\frac{R_s}{t_s} + \frac{R_B}{t_B}\right)^{1/2}$$

V_c — Coefficient of variation of corrected count rate of a "Poisson" distribution.

$$V_c = \frac{\sigma_c}{R_c} \times 100$$

S — Standard deviation of a "normal" distribution.

$$S = \left(\frac{\sum_{i=1}^{n} (X_i - \bar{X})^2}{n-1} \right)^{1/2}$$

Sm — Standard deviation of the mean "normal" distribution.

$$Sm = \frac{S}{(n)^{1/2}}$$

Sp — Standard deviation of a "normal" distribution approximating a "Poisson."

$$S_p = \sqrt{\bar{X}}$$

Total counts accumulated to insure a set value of V.

$$N_S = 20{,}000 \left[\frac{R_S^*}{VR_C^*}\right]^2$$

$$N_B = 20{,}000 \left[\frac{R_B^*}{VR_C^*}\right]^2$$

Counting time required to insure a set value of V.

$$t_S = \frac{20{,}000\ R_S^*}{(VR_C^*)^2}$$

$$t_B = \frac{20{,}000\ R_B^*}{(VR_C^*)^2}$$

t — standard deviation of time for preset count technique.

$t = t/\sqrt{N}$ where N is the preset count value.

$$t = \frac{t\ \text{mean}}{\sqrt{N}}$$

Applications

Data are of little value if the measuring instrument lacks stability. To check stability, 10 consecutive one-minute count rates of a 5 pound KCl source are determined on each of the three pairs of detectors. The standard deviation of the mean σ_c or Sp is determined for each pair of detectors. If there are more than three out of 10 values deviating from the mean by one standard deviation, then instability is to be suspected.

The stability check assumes that the potassium window is used. More counts could be recorded by opening the window to cover a wider energy band, but as the "C" discriminator is lowered, photomultiplier tube noise level begins to go up.

The stability check, when modified, can be utilized when counting subjects. A background count is determined prior to the subject count rate and immediately following it. Three consecutive counts are made on the subject. S and Sp are determined. One of the three trials is expected to have a value different from the mean by one standard deviation, i.e. S. However, it is possible that the background has changed slightly during the counting procedure. If S is greater than Sp, it is suspected that the back-

ground did change.

An example will demonstrate this procedure. The following table presents the data accumulated from three two-minute counts on a human. Background was determined by two two-minute counts.

n	R_B	Rs
1	8930	11030
2		11170
3	8896	10998
$\bar{X}$ =	8913	11066

$(X_1 - \bar{X})$	$(X_1 - \bar{X})^2$
-36	1296
104	10816
-68	4624
	Σ = 16736

$$S = \left(\frac{16736}{2}\right)^{1/2} = 91.48 \text{ cpm}$$

$$Sp = (11066)^{1/2} = 105.21 \text{ cpm}$$

During the six minutes that this person was being counted, the background did not change appreciably. Had S been greater than 105 cpm, the count would have been repeated.

If R_B is subtracted from Rs to get Rc, then the reported standard deviation and the coefficient of variation or error would be:

$$\sigma_c = \left(\frac{11066}{6} + \frac{8913}{4}\right)^{1/2}$$

$$= 63.76 \text{ cpm}$$

$$V_c = \frac{63.76}{2153} \times 100$$

$$= 2.96\%$$

The count rate would be reported as 2153 ± 64 cpm or 2153 ± 2.96%

These counts represent the ^{40}K counts in a human. Coefficients of variation are expected to be 1.9 to 4.9 percent, depending on the size of the subject.

If σ_c is compared to the deviations for the corrected count rates (R_c), two of the three trials should be within ± σ_c. Corrected count rates are 2100, 2257, and 2102, obtained by subtracting the first, average, and second background count rates from the respective three gross count rates on the subject.

The rule for rejecting a count rate and re-evaluating the mean is: If one of the values deviates from the mean by more than *or* less than two standard deviations, then the value should be rejected and a new count rate taken for establishing a new mean. This is known as Chauvenet's Criterion for rejecting a reading.

A count rate is usually considered significant if R_c is greater than $3\sigma_B$. This is the minimum amount the detector is capable of counting, and is an index of precision. The difference between R_S and R_B will be small. The calibration factor is defined as R_C (cps)/A(μC_i). Since $R_S \cong R_B$ then

$$\text{Precision} = \frac{3\sigma_S}{\text{Calibration factor}}$$

where σS is in cps and the precision is in microcuries. The practical value of knowing the precision of a counter is that it tells the order of magnitude of the smallest sample that can be detected. Another term that may be useful is the background equivalent activity. This is the activity of a radioactive material in μC_i which will give a count rate equal to the background.

Background equivalent activity

$$(\mu C_i) = \frac{R_B \text{ (cps)}}{\text{Calibration factor}}$$

A much more eloquent treatment of this topic can be found in other references (5 and 6); however, this will serve the basic needs of the heterogeneous group which will be using the whole body counting facility.

Two types of information can be recorded from the whole body counter: the body's natural radioactive burden of ^{40}K, which has already been treated extensively in this manual, and (2) the bio-application of administered radioactivity utilizing a whole body counter for the collection of data.

Radioactive body burdens from fallout and industrial or research type accidents are usually measured with a crystal whole body counter. It is suggested that a liquid whole body counter can be used as a screening device to estimate burdens in the body. Because of the liquid detector's poor resolution, this method would be followed by a more precise measurement with the crystal detector whenever it was felt necessary. Exploration of this procedure is not a purpose of this manual, however.

Bio-Applications

When there are a large number of radioactive atoms of a particular isotope, the number of atoms that will lose their radioactivity in a certain period of time is predictable. The equation that permits prediction of the number of atoms that remain radioactive is presented by the classical expression

$$A = A_o e^{-\lambda t}$$

where A_o is the original number of atoms present (some use the symbol N_o for number) when the time t is zero, λ is the decay constant for the isotope, e is the base of the natural log (2.7182...), and A is the number of radioactive atoms after some lapse of time t.

The decay constant λ is the fraction of the number of atoms which decay in unit time. The equation is obtained by integration of the differential

$$\Delta A = -\lambda A \Delta t.$$

The negative sign indicates a loss of radioactive atoms. Integration is as follows:

1. $$\lambda = -\frac{\Delta A/A}{\Delta t}$$

2. $$-\lambda A = \frac{\Delta A}{\Delta t}$$

3. $$-\lambda A = \frac{dA}{dt}$$

4. $$-\lambda\, dt = \frac{dA}{A}$$

5. $$-\lambda \int_0^t dt = \int_{A_o}^{A} \frac{dA}{A}$$

6. $$-\lambda t = \ln \frac{A}{A_o}$$

7. $$A = A_o e^{-\lambda t}$$

The time required for a radioisotope to lose 50 percent of its activity by decay is called the radioactive half-life. When

$$-\lambda t = 0.693$$
$$e^{-\lambda t} = 0.5$$
and
$$A = A_o\, 0.5,$$

the relationship between λ and the half-life (T) is

$$\lambda = \frac{0.693}{T}.$$

The time required for the decay of one-half of the atoms in a radioactive isotope is called the physical half-life (T_p). The treatment of biological data demands consideration of two additional half-lives. One is the biological half-life (T_B) and the other is the effective half-life (T_E).

The biological half-life (T_B) is that time required for the body to eliminate one-half of an administered dose of any substance by the regular processes of elimination. The effective half-life (T_E) is the time required for a radioactive element, *fixed* in the tissue of an animal body, to be diminished 50 percent (7). The effective half-life results from the combined action of radioactive decay and biological elimination.

In the effective half-life, T_p and T_B are related by their respective decay constants according to

$$\lambda_E = \lambda_p + \lambda_B.$$

Since $\lambda = \frac{0.693}{T}$, then

$$\frac{0.693}{T_E} = \frac{0.693}{T_p} + \frac{0.693}{T_B}.$$

Reducing the common expression 0.693 to 1, the equation becomes

$$\frac{1}{T_E} = \frac{1}{T_p} + \frac{1}{T_B} \text{ or } T_E = \frac{T_p T_B}{T_B + T_p},$$

and $$\frac{1}{T_B} = \frac{T_p - T_E}{T_E T_p}$$

or $$T_B = \frac{T_E T_p}{T_p - T_E}.$$

The equation for T_B tells us that T_E can never be greater than T_p, i.e.

$$T_E < T_p$$

It also tells us that as T_E becomes smaller, T_B decreases, and as T_E approaches T_p, T_B becomes larger. (By substituting some values for T_E and T_p, this will become more evident.)

Data from whole body counts are usually plotted on semi-log paper. (They can also be plotted arithmetically). The log coordinate can be either count rate (intensity) or fraction (%) of the count rate at time zero. The arithmetic coordinate is some convenient unit of time subsequent to administration of the radioisotope.

Beginning of the count depends on the method of administration. If the material is given intravenously, the count can begin immediately. An oral dose requires some time for partial uptake and mixing. This is also true if given intraperitoneally or intramuscularly. Since many nuclides are eliminated via the urine, it is helpful if the subject urinates prior to entering the counter.

The investigator should always be aware that element toxicity can result if the specific activity of the element is low, since increased amounts of the element would be necessary to produce a given level of activity. Specific activity of an element is defined as the total radioactivity of a given isotope per gram of element (7).

Figure 7-1 demonstrates a typical whole body count. The "A" portion of the curve represents diffusion (mixing) of the material in the body. The "B" portion is organ preferential and hence a reciprocal of the differential uptake time of the different pooling centers within the body. In the "C" portion, body equilibrium, that is, reutilization and excretion at constant rates, has been reached. The "C" part is that period when the material has been "fixed" in the tissues of the animal body as defined by the effective half-life.

Study of the "A" portion of the curve would provide insight into the optimal method of administration of a particular element.

The "B" section presents a picture of the reciprocated rate of uptake.

Finally, exactly how long a material remains in the body, and hence is utilized for the general well being (or detriment, as in the case of radiation exposure), is depicted in the "C" section.

Since T_E is defined only for the "C" portion and is determined by extending linearly the line back to the Y-axis, half-times of the "A" and "B" portions of the curve are not measurements of the effective half-life.

The "A" and "B" portions represent a biological measurement.

The biological half-life for the "C" portion has been defined in terms of T_E and T_p. Since T_E is not defined for the "A" and "B" portions of the curve, the biological half-times can be obtained by correcting the net count rate at times of measurement for radioactive decay and relating this to the percent retention assuming that when t = o, biological and physical decay rates are equal. If R_i is the net count rate for the passage of time t, the net count rate is expected to decrease according to

$$R_i = R_o e^{-\lambda t}$$

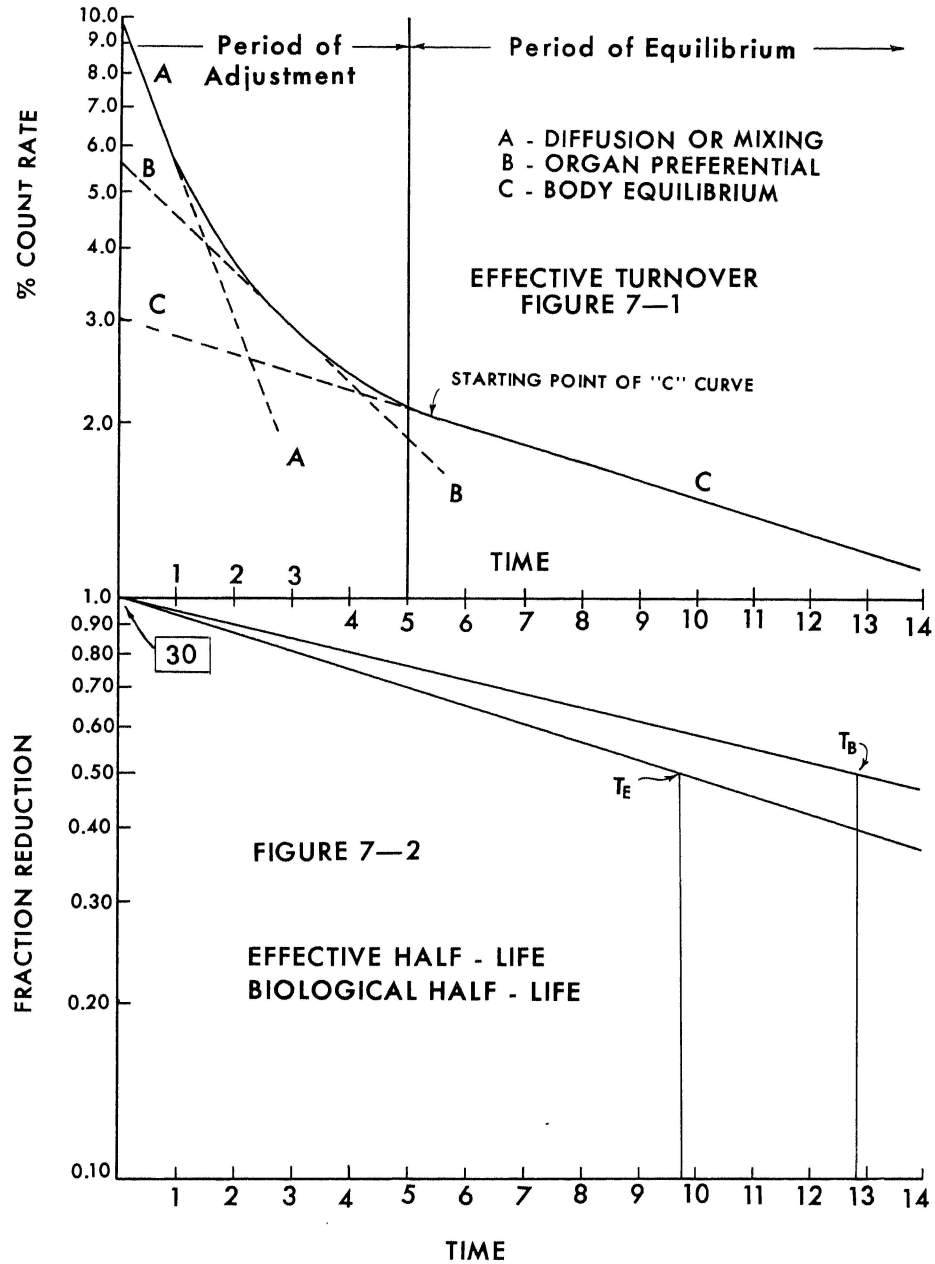
where λ is the physical decay constant, and R_o is the net count rate when t = o. R_i is corrected for decay (R_d) by

$$\frac{R_i}{e^{-\lambda t}} = R_d$$

Values of R_d are divided by R_o and multiplied by 100 to obtain % retention, i.e.

$$\% \text{ Retention} = \frac{R_d}{R_o} \times 100$$

To plot the "C" portion of the effective curve for biological turnover, consider the point where the extrapolated "C" portion crosses the log axis, to be 100%. This can be visualized by replotting the "C"



portion on semilog and allowing the curve to cross the 0.5 (50%) line at T_B. The fraction reduction

$$(e^{-\lambda_B t})$$

can be read directly from this curve for those time periods corresponding to the same time periods on the effective curve.

This is demonstrated in Figure 7-2. In Figure 7-1, the "C" portion crosses the log axis at 30%. The effective half-life is 9.7 and the biological half-life is 12.8. The physical half-life would be 40.

Biological points on the "C" curve are calculated by multiplying 30% by the biological fraction reduction corresponding to the time periods beyond the starting point of the "C" curve. The biological equivalent for time 6 would be 30 x 0.72 or 2.16. For time 10 it would be 30 x 0.58 and for time 13 it would be 30 x 0.49. Thus, the biological turnover of the "C" portion is determined by

$$A = A_o e^{-\lambda_B t}$$

where

$$\lambda_B = \frac{0.693}{T_B}.$$

In this case, A_o is the intercept of "C" with the log axis (30%), t is for time periods beyond time 5.5, λ_B is the biological decay constant, and T_B is the biological half-life.

Another technique for determining biological turnover of an isotopic material is to construct a standard (phantom) identical in size, weight and activity to the specimen and make subject comparisons to this standard. The standard (S_o) is counted each time the subject (S) is counted. The ratio, S/S_o, is plotted on semilog graph paper against time after post administration of the radioactive material. Usually the first two or three days are omitted, since this period represents mixing and is not significant in determining T_B.

The indirect standard method corrects for daily instrument variation and arrives at biological turnover without first determining T_E.

The technique can suffer from some error when utilizing a high efficiency counter such as that used in whole body counting. The exact quantity of activity administered is seldom exactly duplicated in the standard. An error of several hundred atoms of radioactivity in the standard can be corrected by a correction factor, f, established at time of administration.

$$f = S/S_o$$

S_o is the count rate of the subject when time = 0. Thus, the corrected ratio would be

$$S/S_o \times f$$

If daily counts are taken on a large number of animals which are also large in physical size, data processing may become astronomical. Definitely, a computer program and additional labor for handling standards (phantoms) and subjects would be necessary.

Both the internal standard and indirect standard techniques will arrive at the same end point in terms of biological turnover.

The retention curve on semi-log paper is not always curvilinear as suggested; it may be rectilinear. A rectilinear curve is obtained when the isotope is tagged to a biological carrier, for example, ^{131}I-thyroxin or ^{51}Cr-red blood cells.

Miscellaneous

All movable parts and drive mechanisms in the chamber should be lubricated at least once a year. This has been done as a matter of routine in the 1st month of each calendar year. The pots on the *High Voltage Control* panel should be cleaned once a year with silicone contact cleaner. The contact cleaner can be used on the other spectrometer potentiometers. Before moving any settings, make sure they have been recorded. Nothing is more embarrassing than to move a dial setting and then desire to return to that setting only to discover that you have forgotten what the setting was.

Liquid scintillation whole body counters can experience parameter shifts that are not evident in a crystal or plastic type detector. When air or other debris is introduced into a scintillation liquid, a spectral shift down or to the left, and quenching, will occur. Foreign objects in the scintillator will cause it to change color (from clear blue to yellow or orange). Air may be removed by purging the system with ultra high purity nitrogen. Nitrogen is bubbled into the liquid at a very slow rate for 3 to 4 hours. Teflon or glass tubing should be used when purging. Avoid copper, rubber, or other synthetic type tubing. Water or dirt in the liquid demands a change of the scintillator.

Slight pin hole leaks can develop in the thin stainless steel detecting surface of the tanks. The scintillator may also leak around the photomultiplier tube seal and drain pipe joints. Leakage is detected by the prominent odor of toluene in the chamber and located by ultraviolet light. The scintillator will fluoresce upon exposure to the ultraviolet light.

For any repairs or information, contact:
Packard Instrument Corporation
Whole Body Counter Division
2200 Warrenville Road
Downers Grove, Illinois
Area Code 312-969-6000

Whole Body Counting is man's improved effort to look inside the body and observe biological systems collectively. It is nondestructive in method, and, aside from some psychological discomfort associated with being locked up in a steel room and placed under a large detector, offers the investigator a fairly easy technique to examine these systems. The instrument is tempermental, but with some familiarization, operators can achieve results with acceptable limits. Deviation from the established rules for operating the instrument will tend to broaden these limits.

APPENDIX A

Standard Operation Rules for Whole Body Counting

1. Bring all domestic animal experiments in *the east entrance.* Confine animals to the preparation room.

2. Domestic animal researchers and their helpers should confine their activity to the animal preparation room, the east entrance to the counting chamber, and the north side of the building. Unless a change of footwear is made, crossing over into a carpeted area will not be permitted. The animal preparation room floor is concrete. The floor covering of the other areas listed will be vinyl. These floor coverings allow convenient cleaning.

3. Traffic from the Medical Center, Home Economics, and other divisions and visitors enter the south door. Upon entering the building, they are to remove their shoes. The south, the west, and a portion of the east areas of the building have been carpeted for comfort and cleanliness.

4. When using the Whole Body Counter, cover the floor of the counting chamber with a clear plastic cover before starting the experiment. The plastic cover will serve as a catch cover for any manure, dirt, vomit, etc.

5. Smoking is permitted in the animal preparation room and the toilet areas only.

6. Investigators using the facility for a new experiment should review their experiment with the supervisor of the Low Level Radiation Laboratory. A well planned experiment alleviates wasted time.

7. A routine check of the detectors is necessary each day. The earliest time that an experiment can begin is 8:30 a.m. If detector characteristics have changed, deviation from this starting time may be necessary. The researcher will be notified of any changes in starting time. Experiments should not be planned to extend beyond 5:00 p.m. unless special arrangements are made.

8. Adequate help should be provided according to the need of the experiment. Personnel at the laboratory will concentrate on the collection of data and operation of the equipment. Use of their labor for assisting with the handling of the samples will increase the experiment's running time as much as twice its expected time.

9. Researchers are responsible for cleaning the area upon completion of their experiment. This rule is especially important in the animal preparation room where radioactive contamination and disease contamination are possible.

10. Prearrangement for the administration of radioactivity shall be confined to the animal preparation room and the office area in the northwest end of the building. Radioactivity in other areas of the building will not be tolerated. Persons who wish to bring radioactivity to the building must contact Radiation Safety and have their application approved by the Isotope Committee.

11. Going from a carpeted area to a non-carpeted area will generally require a change of footwear.

12. Keep meat samples and cadavers covered with some type of protective covering while they are in the counting chamber. These covers can be reused if care is exercised in their handling and caution is taken to be certain that a soiled undercover is not placed, detector side up, on the next sample admitted.

13. Reactor Facility personnel who have been contaminated and have been requested to report to the Whole Body Counter for gross body contamination survey, enter via the west entrance of the building. Upon entry, remove shoes and proceed to the shower room for possible decontaminating purposes before being counted.

14. These rules shall be completely reviewed by *all* personnel who are actively engaged in an experiment. It is important that everyone realizes the necessity of keeping the building and laboratory as clean as possible.

APPENDIX B

Spectrometer Window Widths

NUCLIDE	*RED*		*GREEN*	
	A	B	A	B
^{40}K			5.00	10.00
^{42}K			5.00	10.00
^{137}Cs	2.50	3.00		
^{131}I	1.30	7.00		
^{64}Cu	1.50	8.00		
^{24}Na	1.50	8.00		
^{59}Fe			3.30	9.50
^{60}Co			3.30	9.50

APPENDIX C

Phantoms

Wt.		*gm K^+*	*Absolute cpm*
lbs.	*kgm.*		
21.5	9.77	475.1	94070
48	21.81	475.1	94070
132	60	1185.15	234660
511	232.27	1411.11	279400
762	346.36	2370.2	469300

APPENDIX D

^{40}K Calculations

1. In 1 gm natural ^{39}K there is
   0.931 gm ^{39}K (93.1%)
   0.060 gm ^{41}K (6.0%)
   0.000118 gm ^{40}K (0.0118%)
2. There are N atoms in 40 gm of ^{40}K where N = 6.025×10^{23}

$$\text{atoms } ^{40}K = 1.18 \times 10^{-4} \text{ gm} \times \frac{6.025 \times 10^{23} \text{ atoms}}{4 \times 10^{1} \text{ gm}}$$

$$= 1.79 \times 10^{18}$$

In 1 gm of ^{39}K there are 1.79×10^{18} atoms ^{40}K.

3. T_p $^{40}K = 1.3 \times 10^{9}$ yr. There are 3.156×10^{7} sec/yr.

$$\text{If } \lambda = \frac{0.693}{T_p} \text{ then } \lambda = \frac{0.693}{1.3 \times 10^{9} \text{ yr} \times 3.156 \times 10^{7} \frac{\text{sec.}}{\text{yr.}}}$$

$\gamma = 1.7 \times 10^{-17}$ sec.$^{-1}$

4. $A = -N\lambda$ if A is in dps* then ...

$$A = 1.79 \times 10^{18} \text{ atom} \times \frac{1.7 \times 10^{-17}}{\text{sec.}}$$

= 30dps

5. $$\text{dpm*} = 30 \text{ dps} \times \frac{60 \text{ sec.}}{\text{min.}}$$

$$= 1800 \text{ dpm}$$

In 1 gm of ^{39}K there are 1800 dpm or 1800 dpm/gm K^+

6. $$\mu C_i = 18 \times 10^{2} \text{ dpm} \times \frac{\mu C_i}{2.22 \times 10^{6} \text{ dpm}}$$

$$= 8.1 \times 10^{-4} \mu C_i$$

In 1 gm ^{39}K there is 8.1×10^{-4} μC_i ^{40}K

8.1×10^{-4} μC_i/gm K^+

7. $^{40}K \xrightarrow[1.32 \text{ mev*}]{\beta^{-}*} {}^{40}Ca$ 89%

$^{40}K \xrightarrow{E.C.*} {}^{40}A* \xrightarrow[1.46 \text{ mev}]{\gamma*} {}^{40}A$ 11%

$$\gamma/\text{min} = 1.8 \times 10^{3} \text{ dpm} \times 1.1 \times 10^{-1}$$

$$= 198 \text{ dpm} \quad \text{or } 198 \text{ cpm}$$

In 1 gm ^{39}K there is 198 cpm* from ^{40}K γ or

198 cpm/gm K^+

3.3 cps*/gm K^+

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* dps = Disintegration per second
β^- = Beta decay
E.C. = Electron Capture
γ = gamma ray
dpm = disintegration per minute
μC_i = microcurie
mev = million electron volts, a unit of energy.
cpm = counts per minute
cps = Counts per second

^{40}K CONSTANT

1.79×10^{18} atoms/gm K^+
$\lambda = 1.7 \times 10^{-17}$ sec^{-1}
30 dps/gm K^+
1800 dpm/gm K^+
8.1×10^{-4} μC_i/gm K^+
198 cpm γ/gm K^+
3.3 cps γ/gm K^+

GRAMS K^+ CALCULATIONS

1. Efficiency $= \frac{\text{net cpm phantom}}{\text{absolute dpm } -\gamma}$ phantom

2. From calibration chart determine Eff. for wt. of subject

3. gm K^+ = net cpm subject x $\frac{\text{dpm} - \gamma}{\text{cpm} - \gamma}$ x $\frac{\text{gm } -K^+}{198 \text{ dpm} - \gamma}$

$$1/\text{Efficiency} = \text{Efficiency}^{-1} = \text{net cpm subject} \times \text{Efficiency}^{-1} \times \frac{\text{gm } K^+}{198 \text{ dpm} -}$$

$$\boxed{\frac{\text{net cpm subject}}{\text{Eff} \times 198}}$$

$$= \frac{\text{net cpm subject}}{\frac{\text{cpm} -\gamma}{\text{dpm} -\gamma} \times \frac{198 \text{ dpm} -\gamma}{\text{gm } K^+}}$$

If Eff. x 198 = k , then $\boxed{\text{gm } K^+ = \frac{\text{net cpm subject}}{k}}$

APPENDIX E

^{137}Cs Determination

Red Channel A = 4.00 B = 8.00 (0.4 - 0.8 mev)
Green Channel C = 5.00 D = 10.00 (1.0 - 2.0 mev)

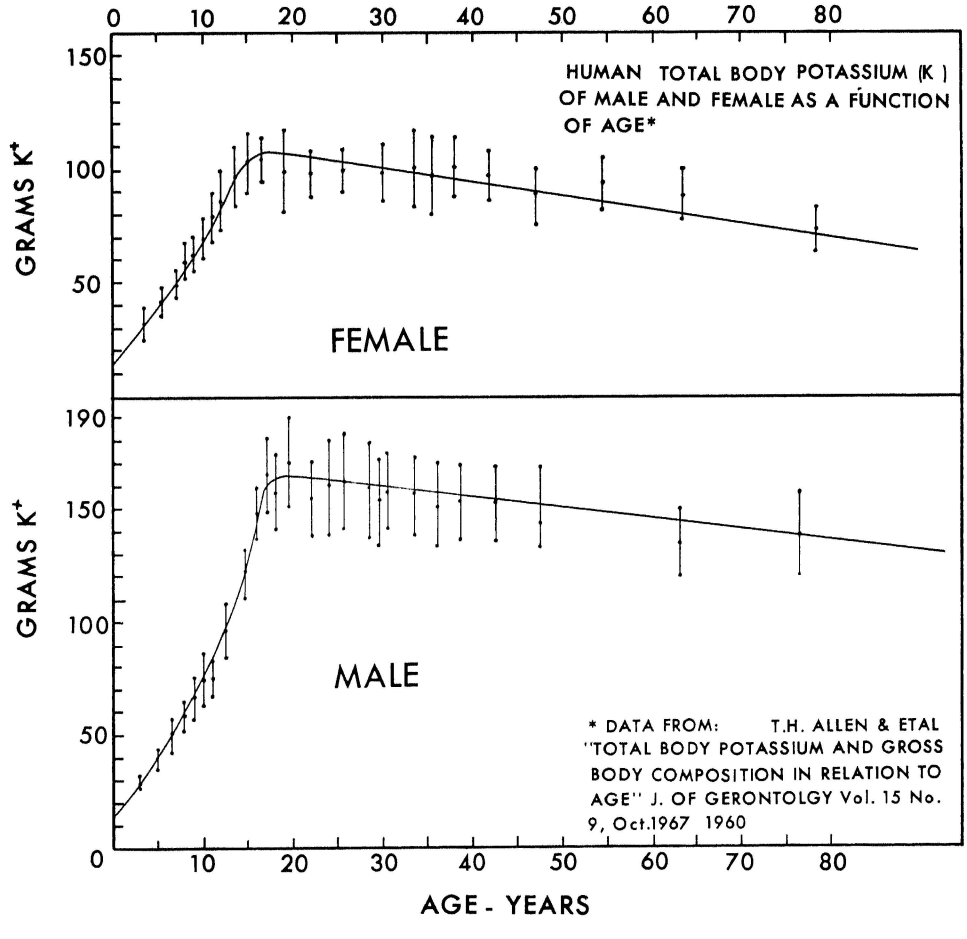
1. With 145.1 lbs ^{40}K phantom determine net count rate in Red Channel (R), and the Green Channel (G).
2. Assuming that there will be an insignificant amount of whatever ^{137}Cs is present spilling over into the Green Channel, determine the ratio.

$$\frac{R}{G} = f$$

which is the fraction of K appearing in Red Channel compared to that which is in Green Channel.

3. Count the subject and determine net count in Red Channel and Green Channel.
4. The product of G and f subtracted from R is the net count rate of ^{137}Cs in Red Channel, i.e.

$$^{137}Cs \text{ (cpm)} = R - (Gxf)$$



References

1. Dubos, R. J., From Remarks Delivered at Biomedical Conference, *Science* 154: No. 3749, 619, 1966.
2. Karzmark, C. J., Photomultiplier Fatigue Effects, *Health Physics* 11: No. 1, 54, 1965.
3. Lohman, T. G., Coffman, W. J., Twardock, A. R., Breidenstein, B. C., and Norton, H. W., Factors Affecting ^{40}K Measurement in The Whole Body and Body Components, *Body Composition Conference,* May 1967. (In Press)
4. Miller, C. E., and Remenchik, A. P., Problems Involved In Accurately Measuring the K Content of the Human Body, *Ann. New York Academy of Science 110:* 175, 1963.
5. Quimby, E. H. and Feitelberg, S., *Radioactive Isotopes In Medicine and Biology—Basic Physics and Instrumentation,* 2nd Edition, Philadelphia, Lea & Febiger, 199-241, 1963.
6. Tittle, C. W., How to Apply Statistics to Nuclear Measurements, *Nuclear Chicago Technical Bulletin No. 14,* 1962.
7. USPHS, *Radiological Health Handbook,* U. S. Department of Commerce Office of Technical Services, 14, 1960.