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Division of Fuel Cycle and Material Safety
Office of Nuclear Material Safety and Safeguards
U.S. Nuclear Regulatory Commission
Washington, D.C. 20555

Dear Sirs:

I am submitting an amendment to the Ohio Wesleyan University license, 34-6535-04, for the use of radioactive materials. This amendment is to allow the use of radioactive tracers in biological research by faculty and students of the Departments of Botany and Bacteriology and Zoology. It is not anticipated that amounts exceeding the unlicensed quantities will be used or stored. However, it will be easier to order radioactive materials with a license. There was a license in the departments in the sixties. The number was 34-6535-2 (064), but the license expired several years ago and the records have been discarded. The present building was built with an isotope room with outside ventilation and other safeguards. This room will be used for storage and handling of the isotopes.

It is assumed that for the first year or so all radioactive work will be under the direct supervision of one of the Mischkes. Since the scintillation counter is presently located at the USDA lab in Delaware, it will be necessary to transport samples for counting. This will limit the use of isotopes. There is a pending NSF grant for a scintillation counter for the university and should that be funded there could be increased use of radioactive isotopes. It is hoped that at that time a radiation safety committee can be formed and the license further updated to show this.

Sincerely,

Charles F. Mischke
Assistant Professor
Department of Botany and Bacteriology

CFM/btb
Enclosures

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4. Use following formulas to calculate present activity of standards in dpm/ml:

For ^3H , ^{32}P , ^{35}S , ^{45}Ca , etc.

$$\text{dpm} = (\text{activity of original standard in dpm/ml}) \times (\text{fraction of activity remaining}) \times (\text{volume in ml of the stock solution})$$

For ^{14}C

$$\text{dpm} = (\text{activity of original standard in dpm/ml}) \times (\text{volume in ml of stock solution})$$

NOTE

The activity of standards for liquid scintillation counting is often reported in microcuries (μC) per milliliter or in microcuries per mole. To convert to dpm, use the following factor:

$$1 \mu\text{C} = 2.22 \times 10^6 \text{ dpm}$$

EXAMPLES

1. Calculation of dpm activity for ^3H on 5-1-75

Original activity of ^3H standard: $2.26 \times 10^6 \text{ dpm/ml}$

Assayed: 6-1-67

Time elapsed since standard was assayed (95 months).

Fraction remaining: 0.639

Amount of standard used: 0.050 ml

$$\text{dpm} = (2.26 \times 10^6) (0.639) (0.050) = 78\,309$$

2. Calculation of dpm activity for ^{14}C on 5-1-75

Original activity: $8.56 \times 10^7 \text{ dpm/ml}$

Stock Solution diluted 1:21

0.01 ml diluted to 0.21 ml

Volume of dilute solution counted: 0.02 ml

$$\text{dpm} = (8.56 \times 10^7) \frac{(0.01)}{(0.21)} (0.02) = 81\,500$$

9.4.3 SAMPLES

To calculate samples:

1. The amount of quench in the samples must be in the range of the standards used to prepare the quench curve.
2. Count samples at the same window and gain settings as the standards.
3. Find the counting efficiency of each sample using the ESCR number of the sample (from the printout) and the quench curve of the isotope being sought.
4. Calculate sample dpm using the following formula:

$$\text{dpm} = \frac{(\text{sample cpm} - \text{background cpm}) \times 100}{\text{percent counting efficiency from quench curve}}$$

SECTION TEN CALIBRATION PROCEDURES

The following procedures will enable the verification of performance specifications using the sealed, argon-saturated, toluene-base standards kit (Beckman 566321 Liquid Scintillation Standards Kit, or equivalent). Should any of the devices not meet a performance specification, refer the problem to qualified service personnel.

10.1 TRITIUM EFFICIENCY VERIFICATION

1. Calibrate gain as directed in Paragraph 7.3.2.
2. Place a ^3H fixed-window Iso-Set or Variable Discriminator Module set for ^3H as directed in Paragraph 6.16, in Channel A.
3. Place a ^{14}C above tritium fixed-window Iso-Set in Channel B, or place a Variable Iso-Set set for ^{14}C as directed in Paragraph 6.16 with LOWER Discriminator set to UPPER setting of ^3H Iso-Set in Channel A.
4. Place PRESET MIN Selector at 20 minutes and PRESET ERROR Selector at 0.2%.
5. Place the sealed unquenched tritium standard in the counting well, depress Channel A Selector Pushbutton, and count for twenty minutes or until 1 000 000 total counts are accumulated ($0.2\% = 2\sigma$).

6. Calculate percent efficiency as follows:

$$\% \text{E (efficiency)} = \frac{\text{CPM (obtained in Step 4)}}{\text{DPM (half-life correction)}} \times 100$$

NOTE

Before determining the percent efficiency, correct standard dpm by multiplying by the decay factor. The decay factor, Figure 28, depends upon the time period between when standard was prepared and when it was used.

The percent efficiency should be equal to or greater than 60%.

10.2 ^{14}C EFFICIENCY ABOVE TRITIUM ENDPOINT VERIFICATION

1. Follow Steps 1 through 4 of the immediately foregoing TRITIUM EFFICIENCY VERIFICATION.

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2. Place a sealed ^{14}C standard in the counting well, depress Channel B Selector Pushbutton, and count for twenty minutes or until 1 000 000 total counts are accumulated ($0.2\% = 2\sigma$). Observe the count rate in Channel B.
3. Calculate percent efficiency as follows:

$$\%E (\text{efficiency}) = \frac{\text{CPM (obtained in Step 2)}}{\text{DPM (from standards label)}} \times 100$$

The percent efficiency should be greater than 65%.

10.3 ^{14}C EFFICIENCY IN FULL WINDOW VERIFICATION

1. Calibrate instrument GAIN Control as directed in Paragraph 7.3.2.
2. Place a ^{14}C full window Iso-Set in Channel B or set a Variable Iso-Set as follows: LOWER Discriminator at zero, UPPER Discriminator at 1000; and place in Channel B receptacle.
3. Place PRESET MIN Selector at 20 minutes and PRESET ERROR Selector at 0.2%.
4. Place a sealed unquenched ^{14}C standard in the counting well and count for 20 minutes or until 1 000 000 total counts are accumulated ($0.2\% = 2\sigma$).
5. Calculate percent efficiency as follows:

$$\%E (\text{efficiency}) = \frac{\text{CPM (obtained in Step 4)}}{\text{DPM (from standards label)}} \times 100$$

The percent efficiency should be greater than 90%.

10.4 SPILLOVER VERIFICATION

A. SPILLOVER IN 50% TRITIUM WINDOW

1. Use the sealed toluene-base tritium and ^{14}C standards provided in the Beckman 566321 Liquid Scintillation Standards Kit.
2. Calibrate the GAIN Control as described in Paragraph 7.3.2.
3. Place a Variable Discriminator Module in Channel A and then complete the following:
 - a. Place ^3H standard in counting chamber.
 - b. Depress Channel A Selector Pushbutton to display count rate of standard on Log Ratemeter.

- c. Set the LOWER Discriminator at zero.
- d. Set the UPPER Discriminator so that the count rate obtained is one-half the disintegration rate of the tritium standard $\frac{\text{DPM}}{2}$. Before determining one-half the disintegration rate from the tritium standard, multiply by the decay factor.
- e. Set the PRESET MIN Selector at 1 minute and count the standard twice to verify the window is set to 50% efficiency, within the statistical limits.
- f. Remove tritium standard from counting well.
4. Set PRESET MIN Selector at 20 minutes and PRESET ERROR Selector at 0.2%.
5. Place ^{14}C standard in counting well and depress COUNT Pushbutton. Observe the count rate in Channel A.
6. Calculate the ^{14}C efficiency (spillover) in the 50% tritium window as follows:

$$\%E (\text{spillover}) = \frac{\text{CPM (obtained in Step 5)}}{\text{DPM (from standards label)}} \times 100$$

The ^{14}C spillover in the 50% tritium efficiency window should be less than 10%.

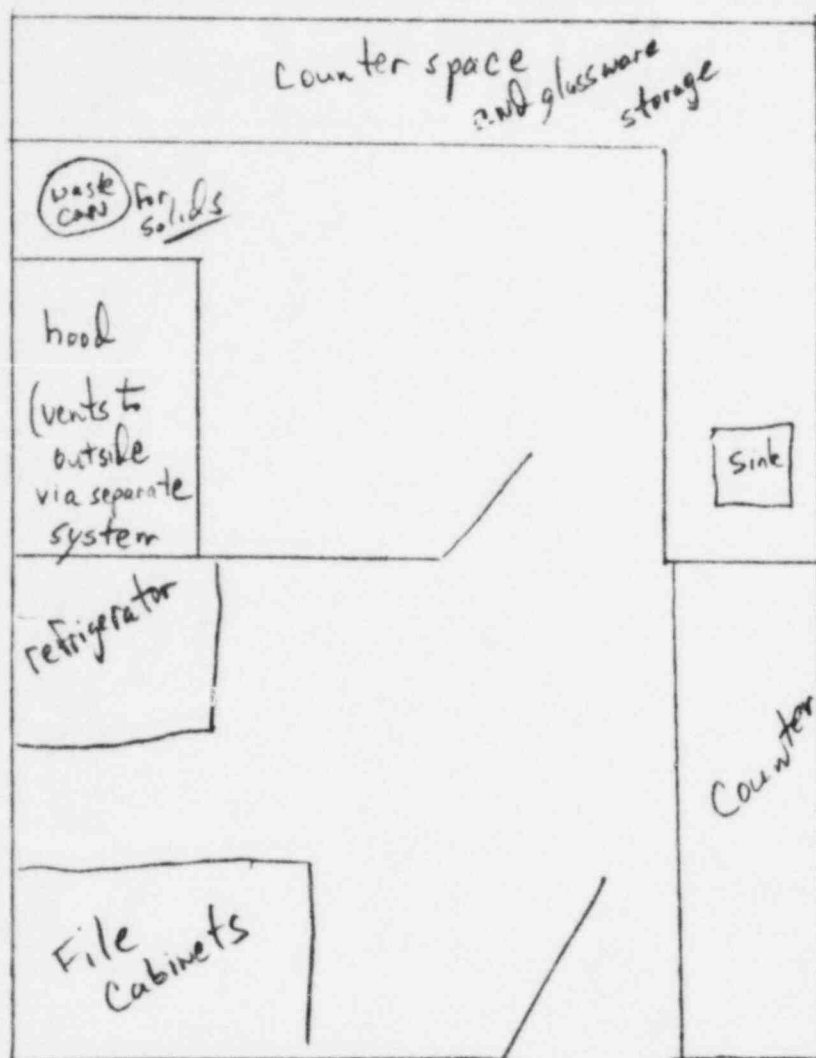
B. SPILLOVER IN NORMAL TRITIUM WINDOW

1. Calibrate GAIN Control as directed in Paragraph 7.3.2.
2. Place a ^3H fixed-window Iso-Set or a Variable Discriminator Module set for normal tritium window as directed in Paragraph 6.16, in Channel A.
3. a. Set PRESET MIN Selector to 20 minutes and PRESET ERROR Selector at 0.2%.
b. Place ^{14}C standard in the counting well and depress COUNT Pushbutton. Observe the count rate in Channel A.
4. Calculate the ^{14}C efficiency (spillover) in the normal tritium window as follows:

$$\%E (\text{spillover}) = \frac{\text{CPM (obtained in Step 3)}}{\text{DPM (from standards label)}} \times 100$$

The ^{14}C spillover in the normal tritium window should be less than 20%.

13. Additional Laboratory facilities for storage and use of isotopes in Bigelow Rice. It is planned that all storage and use should be limited to one or two laboratories in Bigelow-Rice. Below is a diagram of the laboratory designated for such use. It was used for the same purpose from the construction of the building in 1961 to 1970 when the use of isotopes was curtailed.



Room 105 is located in the basement of Bigelow-Rice which houses both the departments of Zoology and Botany/Bacteriology. It is keyed such that only security and personnel using the room have keys.

15. Additions to Radiation Protection Program

- B. Add: Professor Charles F. Mischke as designated to be notified and to receive radioactive material.
- G. Add: General safety instructions will be modified to include Bigelow-Rice 105 in similar manner.
- H. Additional meetings will be held to update the emergency procedures with the use of isotopes in Bigelow-Rice.
- I. (New) Rules for Using Radioisotopes.
See attached sheets. This will be given to all users in Bigelow-Rice and posted in any area where isotopes are used or stored.
- J. (New) Survey Program for Bigelow-Rice.
Since Bigelow-Rice will contain H^3 and C^{14} , it is necessary to set up a monthly survey of areas where isotopes are used or stored. This will involve filter wipes of designated areas and urine samples of personnel who worked directly with isotopes during that month.

CFM/bhd

RULES FOR USING RADIOISOTOPES

Radioisotopes of all types should be considered potentially hazardous. For this reason, we should follow steps to avoid either direct exposure or ingestion of them. Following are some rules that should be followed.

- A. Read safety rules and Nuclear Regulatory Commission manuals for handling isotopes.
- B. Keep accurate records of isotope use and disposal. Use some type of dittoed record sheet.
- C. When handling isotopes, work over a tray at all times and use the special absorbent paper. Use disposable gloves to avoid direct contact and with hard β emitters, always wear a coat and dosimeter and eye shield. Use plenty of paper towels and any transport of isotopes should be made using some type of drip guard (ex: bucket lined with absorbent paper). Sink disposals should be made carefully through a funnel into the sink hole and not the bed. Keep in mind that cleanup of isotopes is very difficult (and sometimes not possible) unless drastic measures are used.
- D. Do not smoke, eat, or keep foods or drinks in areas where isotopes are handled. Never pipet with mouth. Use aids such as pipetors. If materials tend to volatilize (most organics will potentially do this and H^3 is especially bad), use a hood or conduct experiments in well ventilated areas.
- E. Monitor the total work area, including lab benches and floors after each use and keep a record of this, including date. Keep a drawing of lab facilities and indicate where any spillage occurred. The spillage must be made free of contamination. There should be no cpm above BKG that is found in an absolutely unused room. Record your monitoring as cpm above BKG, not as "ok."
 1. For P^{32} , monitoring can be done using a hand monitor, preferably with thin window. The monitor, however, must be calibrated and translatable to mr/hr.
 2. For C^{14} , only a rough idea can be obtained with a hand monitor since the energy is very low. Monitor C^{14} and H^3 using small pieces of wet filter paper. Dry filter and use scintillation counter. Remember, a clean lab has no cpm above BKG found in unused room.
- F. In case of contamination, try to confine spillage to a small area and do not wipe stuff all over. Use materials such as Radiac Wash to help clean up area. Sprinkle concentrated radiac on paper towel and scrub in the spilled area until monitor shows no cpm above BKG. Contact Radiation Safety Officer to report incident.
- G. Isotope disposal
 1. Sink disposal. The NRC lists the amounts that can be disposed into the sink. When sink disposals are made, pour carefully

down hole (with funnel) and not into sink bed itself. Follow with copious amounts of water and monitor sink after use. It should have no cpm above BKG of unused sink.

- a. The amount that can be discarded in sink varies with isotope and type of compound. A fair amount of P^{32} , which has a short half life, can be discarded into sink but much less H^3 or C^{14} can be so discarded.
2. Dry or (separate liquid) disposal. As a rule, let's try to stay well below NRC limits for sink disposal and therefore we must discard most items as dry waste. When possible wash so that most radioactivity is contained in a small volume. If the total volume is small, use paper towels to absorb material and then dispose of these in a plastic bag. If the radioactive material is not volatile but the solvent is, dry in an open beaker at room temperature or low, low heat. Keep in mind, however, that aqueous solutions will be contaminated with bacteria which will transform radioactive materials -- some of which may become volatile. Never use a high temperature hot plate or oven on most C^{14} or H^3 compounds since this would result in contamination of whole lab. If the amount of liquid is large, store in bottles (plastic if aqueous, glass if solvent is toluene).
3. Records:
For liquids disposed into sink, keep a record of amount discarded and time and date of disposal. For dry material, roll up all dirty paper and discardable containers in a plastic bag and place bag inside of cardboard box. Label the box as follows:
 - a. Amount (as mc) placed in box. For the sake of record keeping, for P^{32} and short-lived isotopes, express the mc as amount on basis isotope was monitored at factory. Put date of factory monitoring down. This permits you to avoid decay corrections.
 - b. Type of isotope (ex: P^{32} , C^{14} , etc.). Keep boxes of P^{32} separate from C^{14} , etc.
 - c. As successive amounts of isotopes are added, record this on box and in record book.

For liquids not poured into sink, use recording procedure for dry waste. Keep liquid containing bottles in a sufficiently large metal container so that accidental breakage will not contaminate lab area.

SPECIAL PRECAUTIONS

P^{32} -- This isotope presents hazards due to direct exposure and all precautions should be taken to limit exposure. Use a $\frac{1}{4}$ " plastic shield if there is 10 mc or less and arrange lab so that washings can be done using a shield, if possible. Use carriers made with $\frac{1}{4}$ " plastic for transport. This isotope sticks to glassware and radiac wash soaking for

2-3 days aids considerably in removal. Place radiac in plastic garbage can and place garbage can inside a suitably large metal container.

H³ and C¹⁴. Although direct injury from these isotopes is unlikely, they should be considered highly dangerous if ingested. In addition, these materials can volatilize and thereby contaminate the whole lab. They are difficult to monitor, so take extra care. H³ thymidine is about as bad a material as you can get since it's incorporated into DNA. H³ compounds chemically decay rapidly in freezer and, in many cases, they should be stored in ice box as sterile solutions. Personnel handling H³ and C¹⁴ should submit a monthly urine sample as well as wipes of various areas in the laboratory for counting by the designated radiation safety officer.

All isotopes should be kept in locked refrigerators, cabinets or rooms which are clearly labeled.

CFM/bhc

Item 16 for Charles F. Mischke

<u>Type of training</u>	<u>Location</u>	<u>Duration</u>	<u>On the Job</u>	<u>Courses</u>
a. Principles and practices of radiation protection	Univ. of Arizona	4 yrs	yes	no
	SUNY Geneseo	2 yrs	yes	no
	Denver University	2 yrs	yes	no
	USDA Lab, Delaware	1 yr	yes	no
b. Radioactivity measurement, standardization and monitoring techniques and instruments	Univ. of Arizona	4 yrs	yes	no
	SUNY Geneseo	2 yrs	yes	no
	Denver University	2 yrs	yes	no
	USDA Lab, Delaware	1 yr	yes	no
c. Mathematics and calculations basic to the use and measurement of radioactivity	Univ. of Arizona	4 yrs	yes	no
	SUNY Geneseo	2 yrs	yes	no
	Denver University	2 yrs	yes	no
	USDA Lab, Delaware	1 yr	yes	no
d. Biological effects of radiation	Grinnell College	1/2 yr	no	yes
	Univ. of Arizona	4 yrs	yes	yes
	SUNY Geneseo	2 yrs	yes	no
	Denver Univ.	2 yrs	yes	no

Item 16 for Barbara S. Mischke

<u>Type of training</u>	<u>Location</u>	<u>Duration</u>	<u>On the Job</u>	<u>Courses</u>
a. Principles and practices of radiation protection	Univ. of Arizona	5 yrs	yes	yes
	Michigan Cancer Ctr.	3/4 yr	yes	no
	Univ. of Colorado Medical Center	1 yr	yes	no
b. Radioactivity measurement, standardization and monitoring techniques and instruments	Univ. of Arizona	5 yrs	yes	yes
	Michigan Cancer Ctr.	3/4 yr	yes	no
	Univ. of Colorado Medical Center	1 yr	yes	no
c. Mathematics and calculations basic to the use and measurement of radioactivity	Univ. of Arizona	5 yrs	yes	yes
	Michigan Cancer Ctr.	3/4 yr	yes	no
	Univ. of Colorado Medical Center	1 yr	yes	no
d. Biological effects of radiation	Univ. of Arizona	5 yrs	yes	yes
	Michigan Cancer Ctr.	3/4 yr	yes	no
	Univ. of Colorado Medical Center	1 yr	yes	no

Item 17 for Charles F. Mischke

<u>Isotope</u>	<u>Maximum Amount</u>	<u>Location</u>	<u>Duration</u>	<u>Use</u>
Carbon 14	50 microcuries	SUNY Geneseo	2 yrs	Radioactive plant hormone
	1 millicurie	Denver University	2 yrs	Radioactive amino acids and nucleotides in vitro study
	100 microcuries	USDA-Delaware	1 yr	Radioactive growth regulator uptake by trees
Hydrogen 3	1 millicurie	Univ. of Arizona	3 yrs	Amino acids, nucleotides uptake by plants
	1 millicurie	SUNY Geneseo	2 yrs	Amino acids nucleotides in vitro studies
	10 millicuries	Denver University	2 yrs	Amino acids nucleotides in vitro studies
Sulfur 35	1 millicurie	SUNY Geneseo	3 mos	H ₂ SO ₄ in water Labeling proteins in algae
Phosphorus 32	2 millicuries	SUNY Geneseo	6 mos	H ₃ PO ₄ , labeling nucleic acids in algae

Item 17 for Barbara S. Mischke

<u>Isotope</u>	<u>Maximum Amount</u>	<u>Location</u>	<u>Duration</u>	<u>Use</u>
Carbon 14	1 millicurie	Univ. of Pennsylvania	2 yrs	Radioactive amino or plant hormone incorporation in plant protein
Hydrogen 3	1 millicurie	Univ. of Arizona	2 yrs	Radioactive amino acid incorporation in plant protein
	1.5 millicurie	Michigan Cancer Foundation	9 mos	Radioactive amino acid/nucleotide incorporation in human cell cultures
Phosphorus 32				
	1 millicuries	Univ. of Arizona	3 yrs	H ₃ PO ₄ uptake by algae

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