

SOP TITLE: Labeling of Peptides/Antibodies with I-131

SOP Prepared by: Radu Minea & Emi Eastman

Date Prepared: 1/13/2020

PI: Radu Minea

## SECTION I. PPE AND SURVEY EQUIPMENT

*Itemize required PPE and Survey Equipment*

### PPE:

- Lab coat
- Nitrile gloves (two pairs)
- Closed-toe shoes
- Safety glasses
- Chest and ring dosimeters

### Survey Equipment:

- Ludlum survey meter w/ GM probe

### Additional Protective Equipment:

- Charcoal filtered glovebox hood
- Chemical fume hood
- Lead bricks

## SECTION II. MATERIALS

*Itemize required materials*

- Pierce Pre-Coated Iodination Tubes, 10 borosilicate test tubes (12 × 75mm) coated with 50µg Pierce Iodination Reagent (formerly called "IODO-GEN" Iodination Reagent)
- 1.5mL conical Eppendorf tubes
- Tris Iodination Buffer (25mM Tris•HCl, 0.4M NaCl, pH 7.5). This buffer is used for iodide oxidation and for radioiodination of protein. Store buffer at 4°C.
- Protein stock (50mg/ml) in PBS
- I-124/I-125/I-131 - Iodine-125/Iodine-131 Carrier Free/Reductant Free Radionuclide in 0.1M NaOH (pH 12-14) from PerkinElmer. Shipped ambient in lead pig as Sodium Iodide in 0.1N NaOH as ≈17Ci (629GBq)/mg.

## SECTION III. RADIOACTIVE MATERIAL (RAM)

Isotope	Chemical Form	Supplier	Activity <sub>0</sub> <sup>1</sup>	Activity <sub>1</sub> <sup>2</sup>	Handling Time	Frequency <sup>3</sup>
I-131	0.1M NaOH	Perkin Elmer	2 mCi	2 mCi	2-3 hours	1-2 times/month

<sup>1</sup> Activity<sub>0</sub> = receipt/starting activity    <sup>2</sup> Activity<sub>1</sub> = activity used per experiment    <sup>3</sup> Frequency = frequency of the experiment

## SECTION IV. POTENTIAL HAZARDS & SAFETY PRECAUTIONS

### List potential hazards

- I-131 emits 364 keV gamma rays and 606 keV beta particles.
- The I-131 used in this procedure is volatile. If internal contamination occurs, about 30% is taken up by the thyroid.

### List all safety precautions

The following precautions should be taken while handling I-131:

- Designate area for handling I-131 (and RAM) and clearly label all containers and equipment.
- Line all RAM countertops with absorbent sheets.
- Wear two pairs of gloves at a time.
- Leave survey meter on nearby and frequently survey gloves for contamination. Change gloves as necessary.
- For individuals working with 1 mCi or more of volatile radioiodine, schedule a baseline thyroid bioassay before the experiment and a follow up bioassay within 3 days of the experiment.

## SECTION V. PROCEDURE

### Record steps of the procedure:

1. Before you begin, survey the workspace for contamination. Ensure that everything is at background.
2. Prepare peptide/antibody solution for labeling by aliquoting 75ug of stock peptide/antibody solution into a microfuge (Eppendorf) tube; set aside. Concentration should be >1 mg/ml and in a buffer other than histidine buffer (preferably PBS)
3. Open the lead pig containing the I-131 solution (as Sodium Iodide in 0.1N NaOH) in the fume hood behind a wall of lead bricks and, without taking the screw-cap bottle out of the pig, move the insert plastic barrier on the cap to expose the septum membrane. Using an insulin syringe, inject 200ul of Tris Iodination Buffer (pH 7.5) through the septum into the screw-cap bottle. Place the syringe in a container marked as radioactive waste. Allow 5 minutes for the sodium iodide solution to equilibrate into the Tris Iodination Buffer and trap any free iodine into solution.
4. OXIDATION STEP - Carefully, while handling the screw-cap bottle behind the brick lead wall, unscrew the cap from the bottle containing the radionuclide and transfer with a pipette the entire volume ( $\approx 210\text{ul}$ ) into a pre-coated radioiodination tube; discard the pipette tip into the radioactive waste container.
5. LABELING STEP - Carefully, with the radioiodination tube behind the brick wall, add 10ul from the peptide/antibody stock (representing molar excess of peptide/antibody to available iodine) to the radioiodination tube and then place the rubber lid on the tube. Discard the pipette tip into the radioactive waste container and cap the radioiodination tube. Then manually swirl contents once. Afterwards, gently pulse the Radioiodination Tube every 5 minutes for a total incubation time of 30 minutes.
6. At the end of incubation time, transfer contents of the Radioiodination Tube (i.e., the solution containing the labeled peptide/antibody) into a new Eppendorf and store behind the brick lead wall in the fume hood if used for experiments the same day. For longer term storage, place the Eppendorf containing the radioiodinated peptide/antibody into a makeshift lead sarcophagus and store in a 4°C refrigerator labeled properly with a radioactive sticker.
7. Survey the workspace for contamination. If any contamination is found, discard of the materials or decontaminate.

**SECTION V. PROCEDURE** continued**SECTION VI. SPILL/INCIDENT RESPONSE**

*List instructions for spill/incident response and clean-up. Include notifications to proper authorities (e.g., Rad Safety, DPS).*

- For small spills onto lined countertops, carefully discard of the absorbent lining into the solid RAM waste box. Survey the countertop with a portable survey meter if contamination is suspected. Document the spill and cleanup procedure) used with other radiation records and notify Radiation Safety Services for assistance (323-442-2200).
- For major spills or any personal contamination, contact DPS and Radiation Safety Services for proper instructions and guidance. If possible, contain the spill and check yourself and the area for radioactivity.

**SECTION VII. TRANSPORTATION/STORAGE/DISPOSAL**

*List instructions for transportation, storage, and disposal of RAM*

- Store behind the brick lead wall in the fume hood if used for experiments the same day.
- For longer term storage, place the Eppendorf containing the radioiodinated peptide/antibody into a makeshift lead sarcophagus and store in a 4°C refrigerator labeled properly with a radioactive sticker.
- If transportation of samples is necessary (for example, from one authorized use location to another), place samples in a secondary container to prevent spills onto the floor.
- Store I-131 contaminated objects in sealed containers (e.g. zip-lock bags, plastic containers).
- Do not dispose liquids down the sink. Rather pour into the designated plastic bottles for removal by HazMat.

**SECTION VIII. EMERGENCY RESPONSE**

List instructions for emergency response and notifications to proper authorities (e.g., Rad Safety, DPS).

For emergencies, follow the steps below.

1. Call DPS immediately. UPC (213) 740-4321, HSC (323) 442-1000
2. Call EH&S. Dial (323) 442-2200
3. Provide your name; call back number; nature of the emergency and location.
4. Report the emergency to the supervisor, lab manager, or principal investigator.

**SECTION IX. REFERENCES**

List references

1. *Iodine I-131 Radiological Safety Guidance*. University of Michigan EH&S. <https://ehs.umich.edu/wp-content/uploads/2016/04/Iodine-131.pdf>