Initial Operator Training for the SPECTRAL ami HTX Imaging System with X-Ray Capabilities

This training module is intended to provide initial radiation safety training for individuals who wish to use the SPECTRAL ami HTX Imaging System which has X-ray capabilities. The Imager is located in the animal facility at Molecular Medicine Research Institute (MMRI). The primary use of the ami HTX is to image rats and mice using luminescence, fluorescence, photographic and X-ray techniques. This training module does not include the training required for using open radioisotopes in the radiation laboratory

Training and Exams:

After reading this training module, you must pass a written exam, then schedule hands on training with the MMRI Scientist or RSO. After passing a practical exam, you will be certified to operate the ami HTX Imaging system in X-ray mode.

Responsibilities:

Currently only the MMRI Imaging Scientist and RSO are authorized to use the X-ray function of the imager.

Imaging Scientist: Puja Ravikumar, Ph.D.
pravikumar@mmrx.org
Submit your request for imaging services with Puja.

MMRI Radiation Safety Officer: Jan Rydzewski, Ph.D.
jrydzewski@mmrx.org
510-703-4628
See Jan for any safety or training questions

The following topics will be discussed in this training;

- What are Ionizing Radiation and X-rays?
- How are X-rays produced?
- Radiation Dose
- Health Effects & Regulatory Dose Limit
- General Hazards Associated with the X-Ray imager
- Safety Features and Instructions regarding the ami HTX
- Operating Instructions for the ami HTX

More complete and detailed information regarding the ami HTX Imaging System can be found in the user's manual and other supporting documents located in the imager room or by going to the MMRI Intranet under "Handbooks, Guidelines, Procedures". For access to the MMRI Intranet, use the following links:

Affiliates: http://50.0.117.214

MMRI Employees: http://192.168.1.24

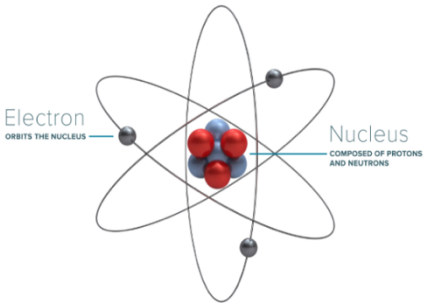
What are X-rays & Ionizing Radiation?

What is radiation?

Radiation is the process by which energy is emitted as either particles or waves. Broadly, it can take the form of sound, heat, or light. However, most people generally use it to refer to radiation from electromagnetic waves, ranging from radio waves, though the visible light spectrum, and up through to gamma waves.

Atoms and Their Parts

Most of the discussion about radiation, how it works, and what its effects are boil down to the interaction of radiation with atoms (and molecules) that it comes into contact with. Atoms form the basic building blocks of all matter. They consist of a nucleus, made of positively-charged protons (and sometimes neutrally-charged neutrons), and an outer cloud of electrons, which have a negative charge. The positive charge of a single proton is equal to the negative charge of a single electron



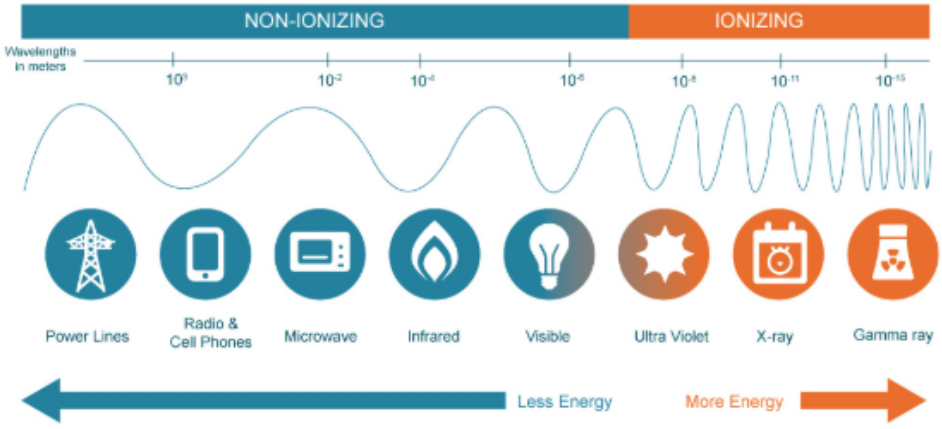
Protons and neutrons have a relatively large size and atomic weight, whereas electrons are extremely small and light by comparison. Due to the nature of opposite charges attracting, atoms tend to have an equal number of protons and electrons, leaving the atom as a whole having a net charge of zero. However, if the atom either loses or gains an electron, it becomes an ion, and carries a charge.

It will seek bonds with other charged particles in order to regain a neutral balance, potentially leading to new molecules being formed.

Ionizing vs Non-Ionizing Radiation

Radiation is generally classified ionizing or non-ionizing, based on whether it has enough energy to knock electrons off atoms that it interacts with, as well as being able to do lower-energy damage such

as breaking chemical bonds in molecules. Ionizing radiation, which is caused by unstable atoms giving off energy to reach a more stable state, is more of a health threat to humans because it involves changing the basic makeup of atoms in cells, and more specifically the DNA molecules inside of cells. It does, of course, take a very strong dose of radiation to substantially damage a cell's structure, as there can be trillions of atoms in a single cell.



The scale of electromagnetic radiation, broken down into categories of ionizing and non-ionizing radiation

Most non-ionizing radiation, such as radio and microwave energy, is considered harmful only to the extent of the amount of heat energy it transfers to whatever it hits. This is, in fact, the way that microwaves cook food. UV light is unique in that while the lower energy UVA and UVB rays are non-ionizing, it does have the capacity to cause harmful effects similar to what ionizing radiation can create, such as an increased risk of cancer due to damage to DNA molecules.

Sources of Ionizing Radiation

Ionizing radiation comes from these main sources:

Natural:

- Radioactive decay of unstable elements in rocks and soil, especially rocks that contain radium and release radon gas
- Cosmic radiation: Nuclear reactions in the Earth's sun and stars in space
- Natural radioactivity in the body: Some radioactive material is ingested with food and water

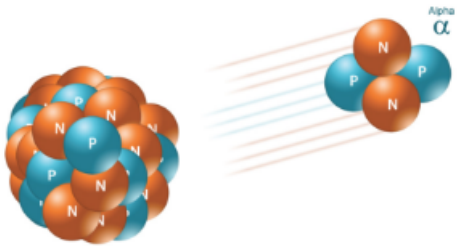
Man Made:

- Nuclear medicine (X-ray machines), radiopharmaceuticals, radiographic sources
- Nuclear power plants, nuclear waste
- Air travel (cosmic radiation)
- Consumer products: smoke detectors, exit signs, luminous watches/clocks

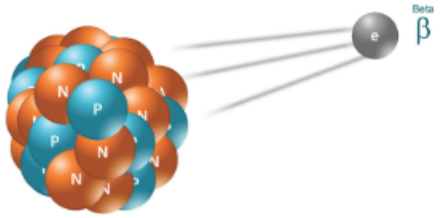
Types of Ionizing Radiation

Ionizing radiation takes a few forms: Alpha, beta, and neutron particles, and gamma and X-rays. All types are caused by unstable atoms, which have either an excess of energy or mass (or both). In order to reach a stable state, they must release that extra energy or mass in the form of radiation.

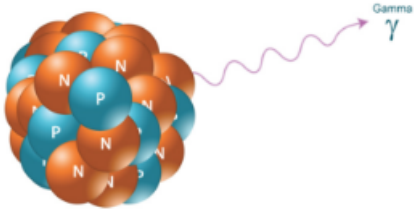
Alpha radiation:
The emission of an alpha particle from the nucleus of an atom



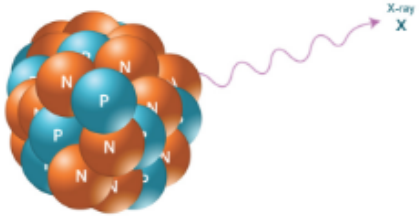
Beta radiation:
The emission of a beta particle from the nucleus of an atom



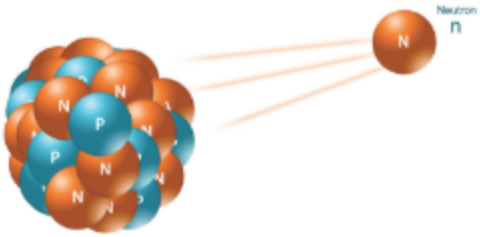
Gamma radiation:
The emission of a high-energy wave from the nucleus of an atom



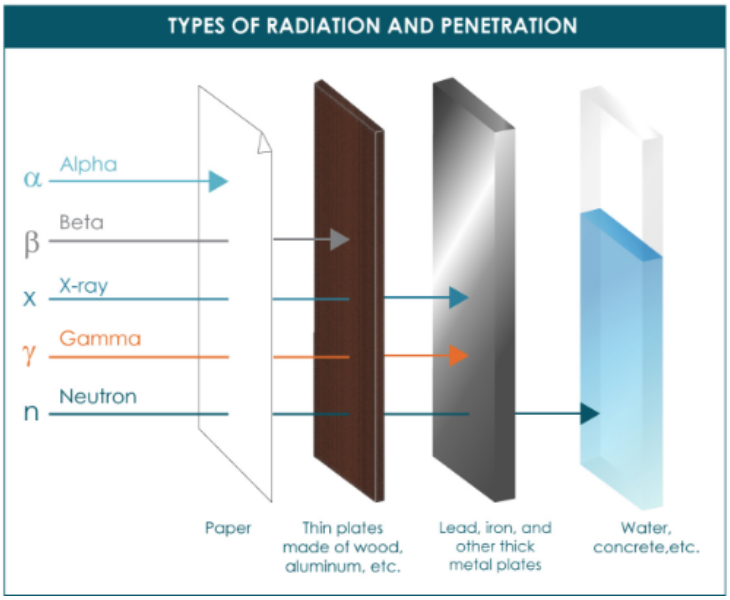
X-Rays:
The emission of a high energy wave from the electron cloud of an atom



Neutron radiation:
The emission of a neutron from the nucleus of an atom



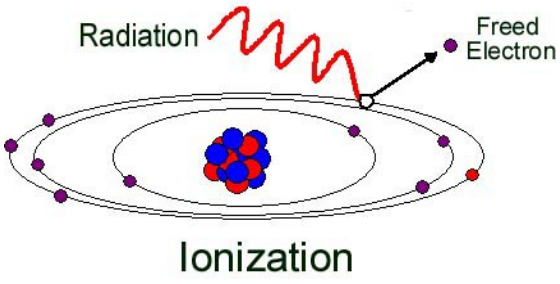
Each type of ionizing radiation can penetrate different materials. As shown in the figure below, alpha and beta particles can be stopped by paper, wood, aluminum and Plexiglas. X-ray and gamma rays can be stopped by lead and iron while neutron emissions can only be stopped by water or concrete. The ami HTX Imager uses steel to prevent all x-rays from escaping the imaging chamber.



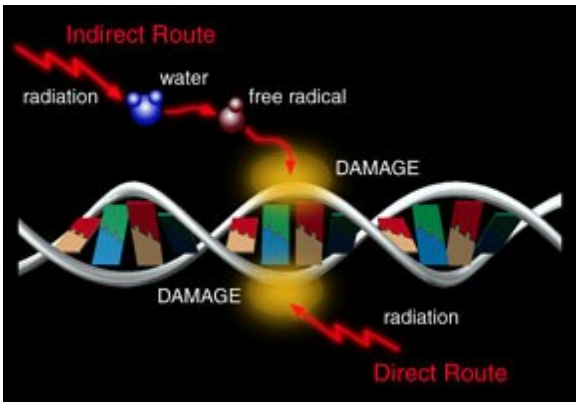
Interaction with Matter and Biological Effects:

When an x-ray or gamma ray is traveling through the human body, it will sometimes interact with an orbital electron of an atom. During the interaction the radiation can impart enough energy to the electron to "free" it from the atom.

The atom now has one less electron, which results in the atom having more positive than negative charges. An atom that does not have the same number of positive & negative charges is called an **ion**. The process of ionization is illustrated on the right.



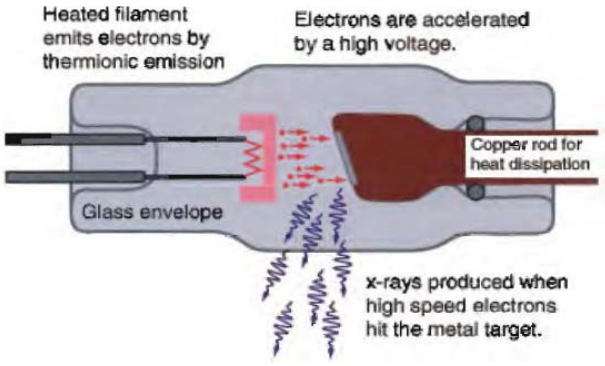
This “freed” electron can potentially cause damage to living cells or DNA. The damage can be repaired from small amounts of radiation, however if enough radiation is imparted, biological damage can occur. The biological damage can include the death of the cell, mutation of the DNA, or preventing the cell from reproducing.



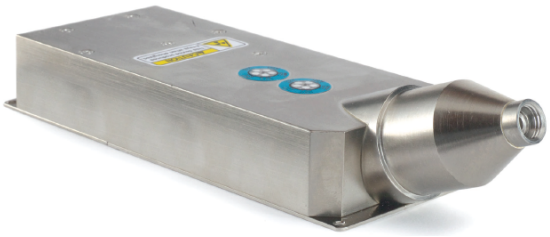
How are X-rays Produced?

The ami HTX uses an x-ray tube to produce x-rays as its ionizing radiation source. It should be noted that the only thing that differs between an x-ray and gamma ray is where they were originated (x-rays are produced outside the nucleus, gamma rays originate inside the nucleus), otherwise you can’t tell the difference between either.

Like the x-ray machine used to take x-rays on patients, the ami HTX Imager uses an x-ray tube to generate x-rays (see picture below). Basically, what happens is that a metal filament is heated up to a very high temperature allowing electrons from the metal to essentially “boil” off. Electrons, the small negatively charged particles orbiting the nucleus, are then accelerated across the glass tube to a positively charged anode. While the majority of the electrons collide with the metal anode and generate heat, some of the electrons lose their energy in the form of x-rays.



X-ray generator used in ami HTX Imager



No radiation is produced in the x-ray tube when the power is turned off.

X-ray Operating Parameters – kV and mA
The energy and quantity of x-rays produced by the x-ray tube is proportional to the operating potential voltage (kVp) and current (mA).

Kilovoltage (kV) determines the penetrating ability (quality) of the X-ray beam. Milliamperage (mA) determines the quantity of X-ray photons. The quantity of radiation remains constant regardless of variations in mA and time as long as their product remains constant

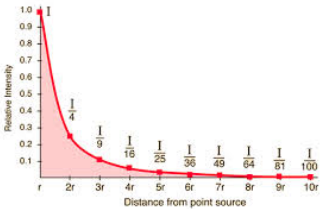
Example: 10mA applied for 1 second = 10mA s = 20mA for 0.5 sec = 10mA s

Radiation Protection Philosophy

The main purpose in the control of radiation exposures are to ensure that any necessary exposures are kept "as low as reasonably achievable**" (ALARA).** Three ways to reduce exposure are:

Time - Decreasing the time spent near a radiation source decreases radiation exposure

Distance – Inverse square law. Doubling the distance from a point gamma or x-ray source, decreases the dose rate by a factor of four. Tripling it decreases dose rate nine-fold



Shielding - Increasing the shielding of a radiation source decreases radiation exposure. Enclose x-ray or gamma sources in dense materials (lead, depleted uranium, or concrete)

Radiation Signs and Warnings

Every X-ray producing machine must have a radiation warning on the unit such as **"CAUTION- Produces X-rays when energized"**. The sign found on the ami HTX Imager is shown below.

Radiation Dose

Exposure

- Amount of radiation traveling through the air. Many radiation monitors measure exposure.
- Units: **Roentgen (R)**

Absorbed Dose

- Amount of energy absorbed by an object or person (that is, the amount of energy that radioactive sources deposit in materials through which they pass)
- CGS Units: **rad (radiation absorbed dose)**
- SI Units: **gray (Gy)**
- 100 rad = 1 gray

Dose Equivalent (effective dose)

- Absorbed dose modified by the ability of the radiation to cause biological damage
- For beta and gamma radiation, the dose equivalent is the same as the absorbed dose. By contrast, the dose equivalent is larger than the absorbed dose for alpha and neutron radiation, because these types of radiation are more damaging to the human body
- CGS Units: **rem (roentgen equivalent man)**
- SI Units: **seivert (Sv)**
- 100 rem = 1 seivert

For practical purposes, 1 R (exposure) = 1 rad (absorbed dose) = 1 rem or 1000 mrem (dose equivalent).

Equivalent Dose Rate

- The rate at which an equivalent dose is received. It is a measure of radiation dose intensity (or strength).
- CGS Units: mrem/sec, rem/hr
- SI Units: mSv/sec, Sv/hr

Total Dose = Dose rate x time

Radiation Exposure and Dose Limits

Everyone on this planet is exposed to some radiation from natural sources and from man-made and medical procedures. For those working with radiation, there are established allowable limits to their annual exposure, which is measured using dosimetry devices.

Average Radiation Exposure to the Public:

Source		Annual Dose (mrem/year)
Natural Background	Terrestrial	22
	Internal Emitters (K-40, U, Th)	34
	Cosmic	28
	Inhaled Radon & Thoron	227
Man-made	Industrial & Occupational	<1
	Consumer Products	13
Medical	Combined	300
	Total	620

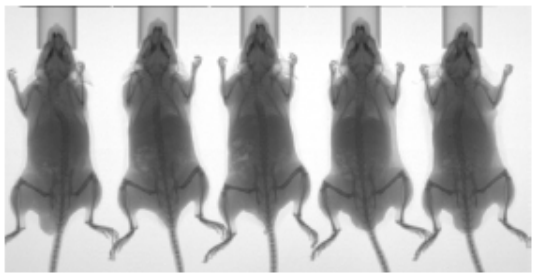
Dose Limits:

	Radiation Worker	Declared Pregnant Worker	General Public
Whole Body	5,000 mrem/year		100 mrem/yr
Lens of the eye	15,000 mrem/year		
Extremities, skin	50,000 mrem/ year		
Embryo/fetus		500 mrem per 9 months	

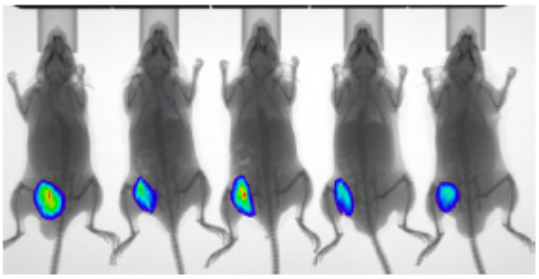
Warnings and Precautions Using the ami HTX Imager

The ami HTX Imaging System includes an x-ray source at the bottom of the instrument. X-rays are emitted from the source through an x-ray transparent window in the metal enclosure. A time delay integration (TDI) x-ray scanner with integrated scintillator plate is located near the top of the instrument. During x-ray imaging, the scanner sweeps above the platform while x-rays are generated for approximately 10 to 15 seconds. X-ray images provide information about internal sample structure and a visual reference when viewing an overlay. The X-ray function can only be activated using a key which is secured by the RSO and the Imager Scientist.

X-ray image



Overlay of fluorescence image on X-ray image.



This unit is capable of producing x-rays with a maximum energy of 40kV. The instrument's steel enclosure blocks all x-rays from leaving the instrument by design, and all units must pass an x-ray leakage test before leaving the factory and again after installation at MMRI, to verify that no x-rays escape the imaging chamber. Manufacturers are required to certify that their products meet the Federal radiation safety performance standard for cabinet x-ray systems. Specifically, the standard requires that the radiation emitted from a cabinet x-ray system not exceed an exposure of 0.5 mR in one hour at any point five centimeters from the external surface. This done by slowly moving a radiation monitoring device over the machine at a distance of 5 centimeters. For comparison, the average person in the United States receives a dose of about 360 mrem of radiation per year from background radiation. Note that 1 mR of exposure to x-rays will result in approximately 1 mrem of dose. Individuals using the ami HTX are not required to wear radiation badges to monitor exposure.

Federal standard also requires safety features that include warning lights, warning labels, and interlocks.

Hardware safety interlocks in the ami HTX prevent the x-ray source from being turned on when:

- The imaging system door is open
- The x-ray key is not in the lock or is turned to the “OFF” position
- The emergency stop button is depressed
- The cable access port cover is not installed (on systems which have the optional cable access port feature)

No x-rays will be produced under any of the above conditions.
Thus, in order to acquire an x-ray image, the door must be closed, the x-ray key must be turned to the ON position, the red emergency stop button must be disengaged (the button must be in the “reset” position by turning the button as indicated in the figure below) and the cable access port cover must be installed. The emergency stop will immediately turn off power to the X-ray source.

ami HTX Imager operating controls and their function:

SPECTRAL ami HTX Front Panel

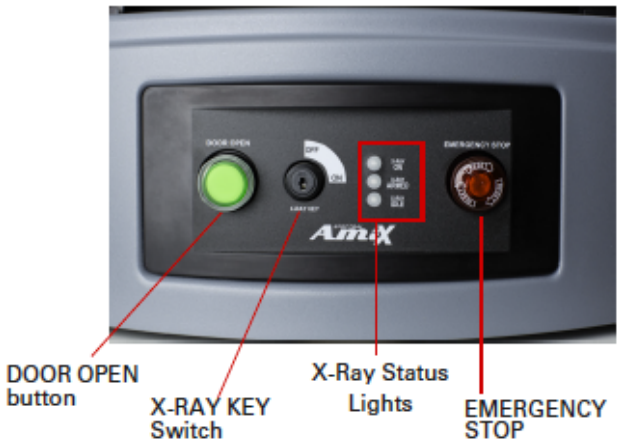
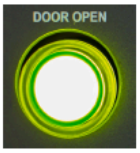


Table 4.6 User-Accessible Operating Controls

Operating Control		Description
DOOR OPEN		Illuminated green push button located on the lower front of the imaging system. Push the button when it is illuminated to open the door. The button does not illuminate during image acquisition.
X-RAY KEY ami HTX Imaging System only		The X-RAY KEY switch is located to the right of the door open button and must be turned to the ON position using the x-ray key to enable X-rays. The key must be turned to the OFF position before it can be removed.
EMERGENCY STOP		Illuminated red button on the lower front of the imaging system. Pressing this button turns off power to the X-ray source (ami HTX Imaging System only) and all motors. It must be reset by turning the button fully clockwise. If the EMERGENCY STOP button is pressed while aura software is running, the software must be restarted before imaging can resume. When the ami HT or ami HTX Imaging System is powered on, the EMERGENCY STOP button will blink if it was pressed and reset is required.
ON/OFF power switch		Located at the top rear of the ami HT or ami HTX Imaging System (see Figure 4.9 on page 55). Energizes or de-energizes the imaging system. This switch controls power for the ami HT or ami HTX only. The computer, monitor, and anesthesia pump system (if applicable) each have their own ON/OFF power switch.

WARNING! Do not attempt to defeat any of the safety interlocks.

X-ray power on ami HTX Imaging System

- Low power: 35 kV tube voltage and 175uA tube current
- High Power: 40kV tube voltage and 200uA tube current
- Resolution: Binning level for the X-ray image. High resolution= no binning, low resolution 2 x 2 binning
- X-ray exposure time: 10 - 15 sec

Radiation Exposure

As previously discussed, there is no radiation exposure unless the unit itself is compromised. Examples of unsafe conditions which could result in potentially dangerous radiation levels include;

- Door interlock to the ami HTX Imager does not work
- Shielding has been damaged
- Evidence of machine tampering

Other Precautions

- No unauthorized personnel may defeat or override any safety features on the x-ray generator or shielding without permission of the manufacturer (Spectral Instruments). The ami HTX is to be serviced by trained personnel only.
- Please keep unit dry.
- Clean the outside surfaces of the ami HTX using a lint-free wipe and soap and water or any of these approved cleaning solutions:
  - 70% alcohol (ethyl or methyl)/30 deionized water
  - Clidox®-S
  - Sporicidin®
  - Trifectant® Disinfectant
- To clean the inside the ami HTX, do not spray or pour liquids directly into the imaging chamber. Saturate a lint-free wipe first then gently clean the platform or walls. Due to potential fluorescence properties of some cleaning solutions it is recommended to wipe surfaces with deionized water afterwards taking care not to allow water to pool on the platform
- Do not use the top of the unit as a storage area. Do not place any heavy items or items containing liquids or materials that may harm the unit if they leaked or spilled on top or inside

How to Use the ami HTX Imager

Note:
Currently only MMRI RSO (Jan Rydzewski, Ph.D.) and the Imager Scientist (Puja Ravikumar, Ph.D) are permitted to use the X-ray function of the ami HTX Imager.

In this section the basic operation of the ami HTX Imager will be discussed. The procedure for basic operation is posted on the unit.

> **WARNING**
> Any use of the ami HTX Imager not for its intended use may result in an unsafe condition. Do NOT insert any flammable or potentially explosive materials into the unit, or apply toxic or corrosive chemicals.
>
> If you have any questions about its use, please refer to the Operating Manual (located in Imaging room) or contact MMRI Imager Scientist

Getting Started/Prerequisites

- To use the ami HTX Imager an individual must complete initial training which consists of completing this training module and perform a hands-on training with the MMRI Imager Scientist.
- Imaging sessions using the ami HTX must be reserved and are scheduled with MMRI Imaging Scientist

Prepare Animals for imaging

1. Please sign in the user log sheet before starting.
2. Animals can be dosed with substrates like D-Luciferin in the imaging room prior to the session.
3. Place the animal in the anesthesia induction chamber and adjust the oxygen flowmeter to approximately 0.5-1.0 L/min.
4. Adjust the isoflurane vaporizer to approximately 3% for induction and 2.5-3% for maintenance of anesthesia.
5. Before placing the animals in the imager ensure that isoflurane is available in the individual nose caps in the ami HTX imaging chamber and the animals are asleep.

6. Close the Imager door and begin imaging by logging in your company user ID.

7. Ami HTX imager has a provision for maintaining heated platform (37°C) to avoid hypothermia in animals. Remember to keep the platform heating in “ON” position.

Recording an X-ray Image

1. Use the key to turn the power on/off switch to on (the key to power on the unit should remain inserted when X-ray function is in use).

2. Select the acquisition modes : Fluorescence, Luminescence and X-ray at the left end of acquisition control areas. Ami HTX can acquire images in all three modes.

3. Check the “photograph” box if need to collect a photograph along with other acquisition modes.

4. If using bioluminescence along with X-ray, select X-ray “ON” in order to illuminate your animals on the platform with X-rays during a luminescence exposure (i.e. X-ray induced optical luminescence image).

5. If using fluorescence along with X-ray select the desired excitation and emission wavelengths along with the exposure time.

6. Select High or low power for X-ray

7. Ensure the X-ray key is turned ON

8. Click acquire and save your image in your folder

9. Please export all your images from the ami HTX desktop within 2-3 days of imaging using the USB flash drive.

10. For details please refer to chapter 3 : Image Acquisition in the Ami HTX instruction manual.