Analytical Technologies in Biotechnology

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Module - 2
Radioisotopes Techniques
Lecture - 2
Basic Concepts 2

Previous lecture, we have discussed about the basic concepts of radioisotope technique. If you could recall, we discussed about the nature of radioactivity, where we have discussed about the atomic stability. Now, if you, as in atomic stability, the ratio of neutrons to protons determine and with various nuclear forces, determine whether a nucleus will be stable or it will be unstable.

Now, when it is unstable then that nucleus is bound to emit either radiation or matter or both to become stable, in this process, it might change its characteristic that is the element can change from one form to another, the process is known as transmutation. Now, this decay could be occurring for several reasons, one there could be a neutron to proton ratio could be high or neutron to proton ratio may be too low and other factors.

This decay, which is in terms of electromagnetic radiation or subatomic particles, can happen through various ways. Like for example, like we have discussed earlier, the decay could happen by emission of a positively charged or negatively charged beta particle, which is called negatron, which is negatively charged and positron which is positively charged. It could occur by heavier nuclei, which is called alpha particle or helium nucleus or it could occur by a simple electromagnetic radiation, which is gamma rays.

In one case we have discussed electron capture where x rays are generated. So, there could be different ways where a radioisotope can decay. Now, in this lecture we will further take our discussion and understand other concepts in these radioisotope techniques. Now, if you could recall we ended the discussion in the last lecture, we were discussing about decay energy.

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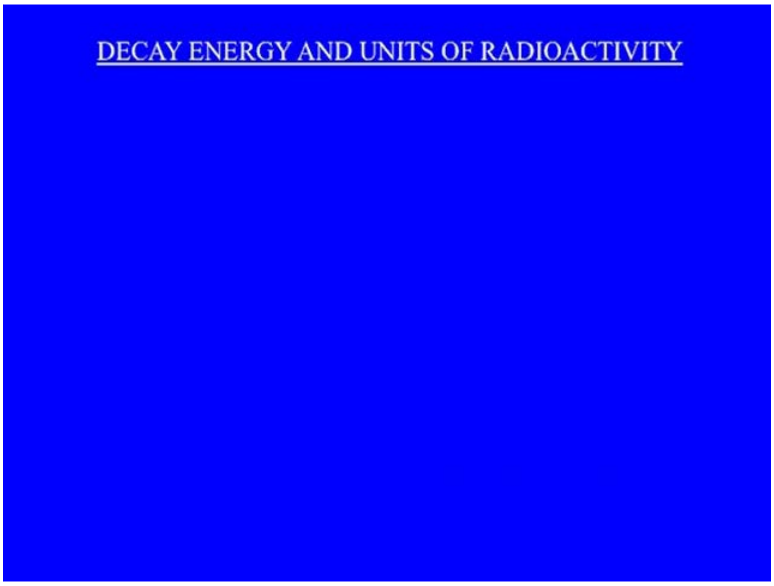

Now, we will start with decay energy and units of radioactivity. Decay energy like I said, they are mostly expressed in terms of electron volt. But, 1 electron volt is not large enough energy and so the majority of isotopes, the term million or mega electron volt is more applicable. Now, if you compare different emitters like alpha emission particle or beta particle or gamma radiations, their energies have particular characteristics. For example, alpha particle alpha emitters have a decay energy in a range of 4 to 8 million electron volts, while beta and gamma emitters in general have weaker energy less than 3 million electron volt. Now, let us little, go more into beta and gamma.

Gamma also have a range, where it is up from ten thousand electron volt to ten million electron volt. But, for beta emitters there is a range of energy and this particular range of energy is called beta spectrum. Because, you have from very low energy, that is a continuous range from very low energy to very high energy, the beta particles are emitted.

This is if you plot it, this is a relative probability of emission of a beta particle, as a function of energy then it is called a beta spectro. Let me show you this on your screen that how different beta particles or beta emitters have a range of energy where beta particles are emitted. Now, let us take a few examples here, where we can see the energy spectrum of negatron emission.

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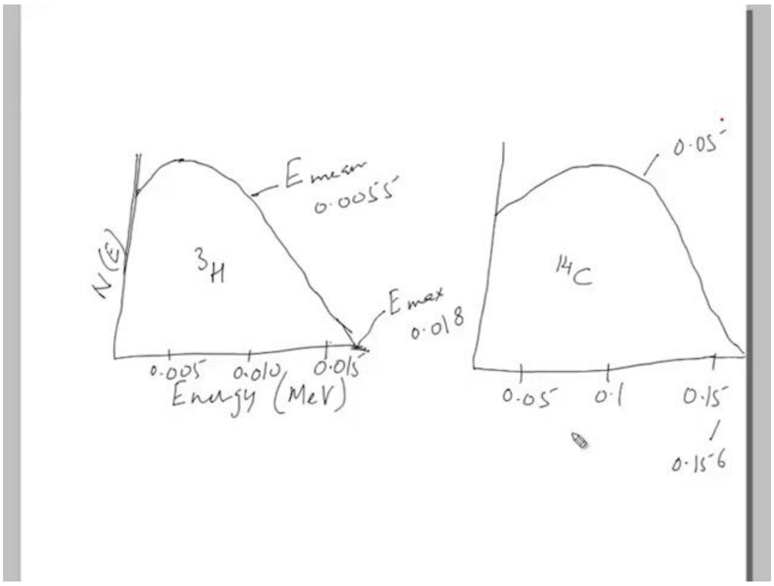

Now, for example if I draw this for few of those negatron emitters, now this specifies N is the number of atoms with a particular energy E and this is energy in terms of million electron volt. So, if I just plot for example, as you will see then if I am plotting for H that is 3 H, then this the plot looks something, it is a very approximate plot, so you have to understand that it will look something like this. Actually, where this will be E maximum and which comes around 0.018 million electrons volt. Now, remember here number of atoms with that particular energy are very less or almost none.

Here, number of atoms with low energy they are very high, so we take somewhere around a mean, that is number of atoms with a mean energy and that comes around 0.0055 million electron volt. So, this is like you can see the beta spectrum for 3 H. Now, if you compare this to others like for example, let us see for another beta emitter that is a medium energy beta emitter 14 C. And these excess will be same as we have drawn here that is N E, a number of atoms particular energy and energy in terms of million electron volt, then it will be some where it will also show a particular characteristic pattern.

Now, here it is having little higher energy because 3 H is a weak emitter, but you are what you called 14 C is the medium energy emitter. And this particular, here if you see the E maximum is around 0.156, this E maximum here is around 0.156 million electron volt and e mean. If I have to say that is around 0.05 million electron volt its higher than the 3 H, but there is a spectrum here in where you will find that less and less number of atoms are at high energy and so you can have a mean value.

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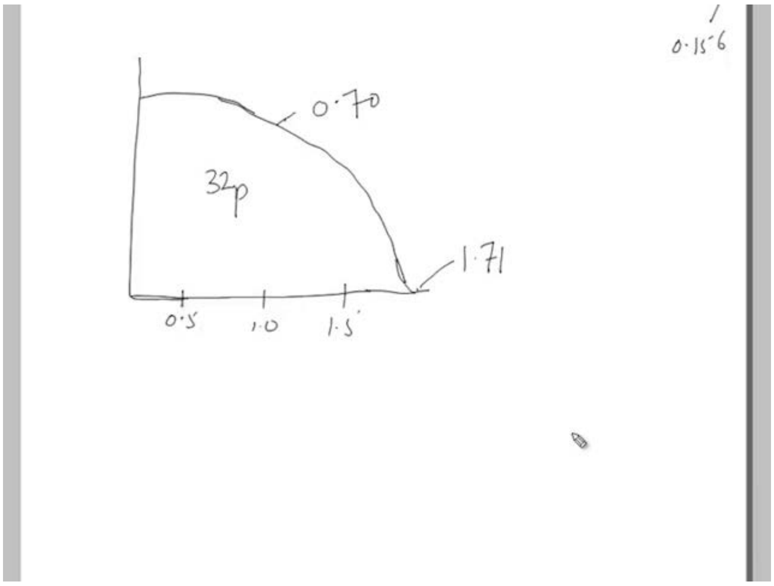

One more example I can take here and that is of particular one which is 32 p I think all of you are aware about it, so it has it is you can say strong emitter and this has higher energy of particles. So, if I say this it has a higher energy and it is rather a different one. Now, here E maximum is almost 1.71 and E mean is somewhere around 0.70. You can see the beta spectrum here that they are being emitted at from zero to a maximum energy. So, his is characteristic feature of only negatron diameters and not the alpha or the gamma radiations. Actually, they have a particular range, so I hope you have been able to understand that what is, what we mean when say beta spectrum in here, all right?

So, let us move on now, all right? So, this was like how a particular beta emitters, emits particles in a continuous range of energy from zero to a maximum value and we take E max for this particles which is near the mean energy and it is roughly one third of E max, all right? So, as we have seen in the graph or a spectrum the particles with E max are very less and particles somewhere in between are taken as number, all right? Now, how this radioactivity in terms of units is expressed, let us see that. An international system of units that is SI system, the units of radioactivity is Becquerel in honor of the scientist Henri Becquerel.

Now, 1 Becquerel is defined as one transformation or disintegration per second and it is mostly expressed in terms of giga Becquerel that is 1 into 10 is to power 9 d k per second or terra Becquerel, that is 1 into 10 is to the power of 12 d k per second and its commonly used, these are the are most commonly used. There is another widely used unit, which is Curie based on Madame Curie and this is expressed as Ci. Now, 1 Ci or one Curie is defined as, number of disintegrations per second per gram of radium and it equal 3.7 into 10 is to the power 10 disintegrations per second. All we can say 37 giga Becquerel. Now, for most biological applications the unit is too large and micro Curie or mille Curie is used rather than simply the Curie.

So, for practical purposes it is not possible to detect actual number of disintegrations, so we have to keep in mind when we are detecting and as we would discuss in coming lectures this is very difficult to capture or to like record actual number of disintegrations taking place. So, what you really count is the number radiations you have counted and it depends on efficiency of counting that how many particular events radioactive events, which you can count. Therefore, radioactivity is generally a stated as detected detection or detected counts per second for example, it could be c p s or it could be counts per minute.

For a particular quantitative experiments, where you require absolute amount of radioactive material to be calculated, then this value that is counts per minute or counts per second needs to be converted into disintegration per minute or disintegration per second. This is done by dividing the efficiency of counting does this, so that is methods to detect determine absolute counting.

Now, in most experiments radioisotopes are really pure actually, that means a carrier of the stable isotope of the element is added in here for stability in other factors. Therefore, the amount of radioisotope present is expressed in terms of a specific activity that is either disintegration per minute or counts per minute or Curie, whatever you are taking, unit per unit mass in terms of moles or grams. You can say counts per minute per gram or per mole, so that is the specific activity you take.

An alternative method of expressing a specific activity, not so frequently used is atom percent accesses, which is defined as number of radioactive atoms per total of hundred atoms of the compound. But, that is not so frequently used, so what you have is one thing is that you have, if you want to do absolute counting, you have to convert counts per minute into disintegration per minute, then you have to also consider about specific activity as there is a stable carrier of the in terms of a stable isotope being mixed with a radioisotope. So, these are important things.

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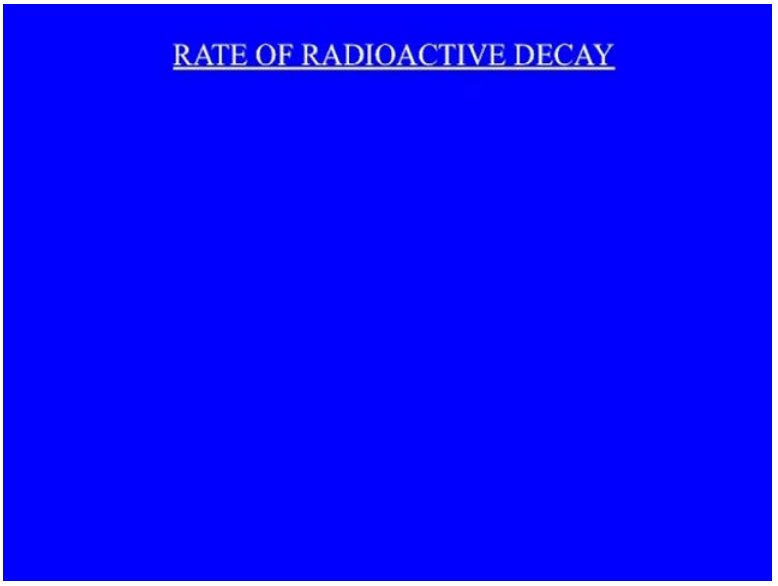

Let us move on into rate of radioactive decay. Now, rate of radioactive, we have seen what are the units being used, how it is expressed in terms of energy. Now, let us get into that. How do you measure rate of radioactive decay? Now, the emission is very spontaneous process, like I said earlier its decay occurs, because of processes, which are confined, to the nucleus of unstable atom and without any physical interaction from outside the atom. So, this is a spontaneous process. Another important part is that it is a random process, radioactive decay is a random process at a single atom level.

What does that mean? It means that as per quantum theory, it is not possible to predict when a particular nuclei will decay. The decay of a particular nucleus will not in any way affect the time of decay of other nucleus, so it is a totally, it is a completely random process and it is not possible to predict when an atom is going to decay. However, the chance that a particular atom will decay is constant over time. So, this decay rate for the collection of large number of atoms is predictable and it can be calculated from the major decay constant of the nuclide. So, this could be done for a group of atoms or large number of atoms, but not for individual atoms.

Now, let us understand this the number of atoms of radioactive material disintegrating per unit time, is proportional to the number of atoms of the isotope present at that time. So, if you have n number of isotopes present at a particular time, then the rate of disintegration will be proportional to that number and it will like as you go along, it will depend on number of radioisotopes present at that particular moment.

So, what does that mean for a given sample of a particular isotope? The number of decay events will be expected to occur in a small interval of time and they will be proportional to the number of atoms present. That is, if you can see here that is the dN upon dt is proportional to N, that is original number of radioisotopes present at that time.

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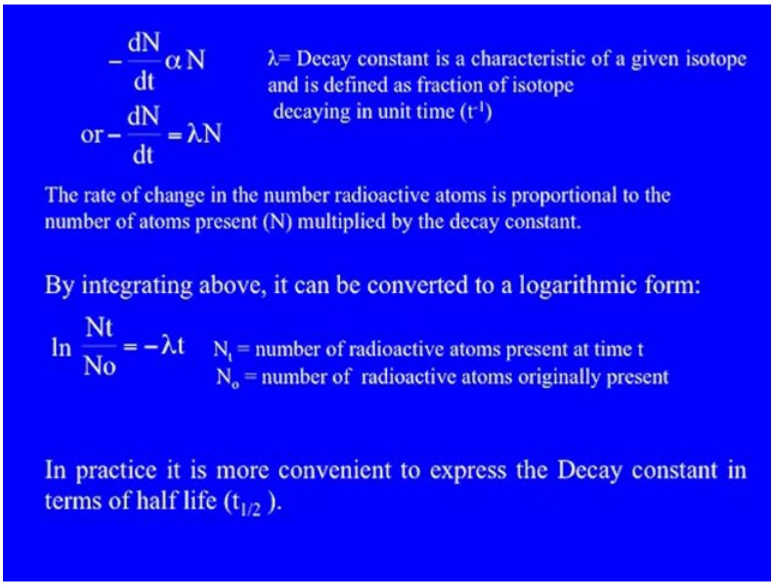

Now, this could be converted to d N upon d t lamda N, where this particular quantity is known as decay constant and it is a characteristic of a given isotope and it is defined as fraction of isotopes decaying in unit time. So, the rate of change of the number of active atoms proportional to the number of atoms present, multiplied by the decay constant. Now, this decay constant is a characteristic of each of the radioisotopes, now if you can integrate her and convert it into algorithm form.

The N t upon No is N t is the number of radioactive atoms present at time t and No is the number of radioactive atoms originally present is equal to lamda t. Now, it is much more easier. Now, here to express the decay constants in terms of half-life, we will see what is half-life and we will discuss that.

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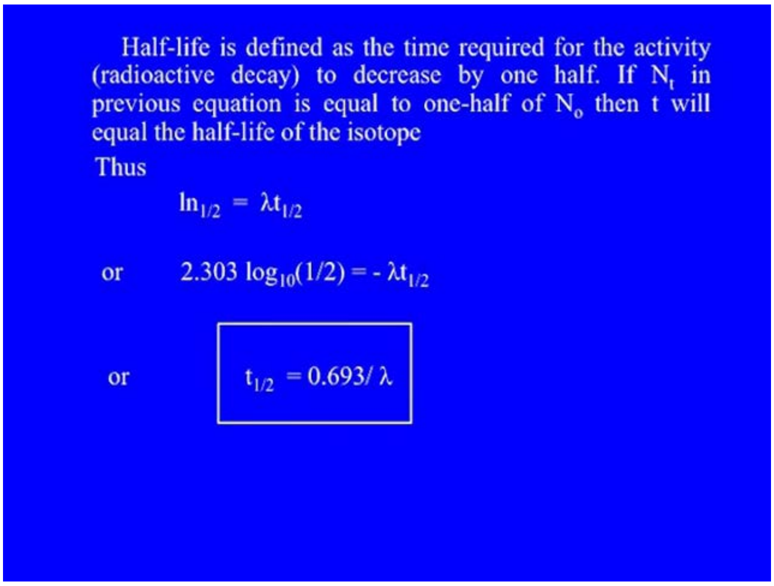

Now, half-life will be defined as the time required for the activity or radioactive to decrease by one half, so if N in the previous equation is equal to one half of N zero then t will be equal to the half-life of the isotope. S shown here, in this that you have taken the log logarithmic value and then finally, what you get is t half that 0.693 upon lamda which is the radioactive decay constant.

So, what you can get is that you have particular, you can express the rate of radioactive decay in terms of half-life that is that how much time does it take to decrease by one half of the original value. We will see it is a exponential quantity that is there is a exponential decrease. Here, now values of t half can vary widely from almost like 10 is to the power 19 years for the lead 2 0 4 to 3 into 10 is to the power minus 7 seconds for polonium, so it could very large or it could be very small actually, all right? Let us little bit discuss about half-life.

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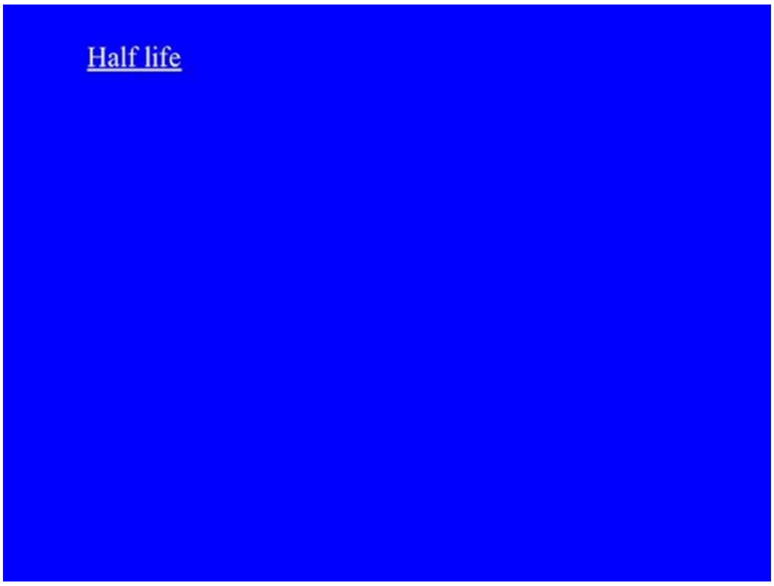

So, half-life is abbreviated as t half and half-life is the period of time, it takes form the amount of a particular substance undergoing decay to decrease by half. Now, half-life is used to describe quantities undergoing exponential decay. For example, radioactive decay where the half-life is constant over the whole life, at the decay. Now, it is the characteristic unit for the exponential decay quotient, like I told you and it is expressed in that form.

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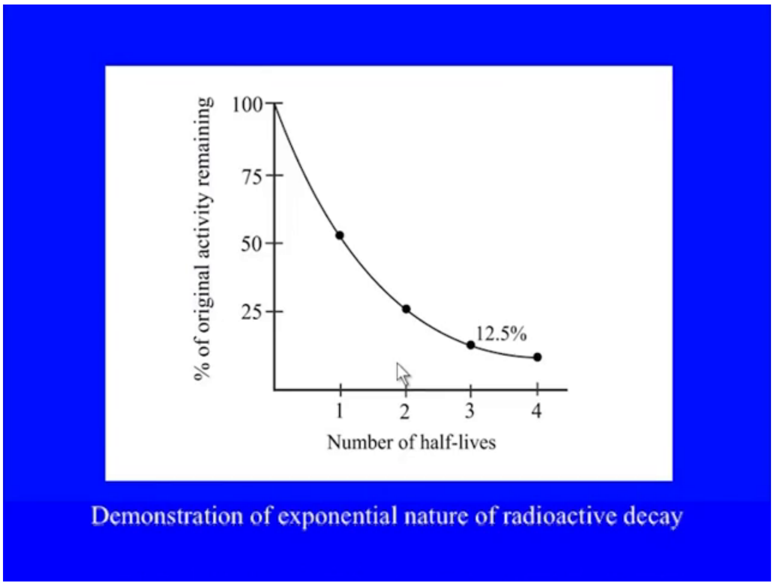

If you see this figure here, this shows demonstrates the exponential nature of the radioactive decay. Now, here if you see there are number of half-life’s given here, like half-life one, which is at 50 percent, half-life two, which will be 25 percent, because original value becomes 50. So, it is now 25 percent and that half-life three will come what it will be? 12.5 percent, because it is half of 25 and likewise you can keep on going.

So, a particular radioisotopes decays in terms of half-life, as we calculate the rate of decay actually and it is an exponential nature of radioactive decay, so you can calculate from the original event that how many half lives particular radioisotopes have gone through, all right?

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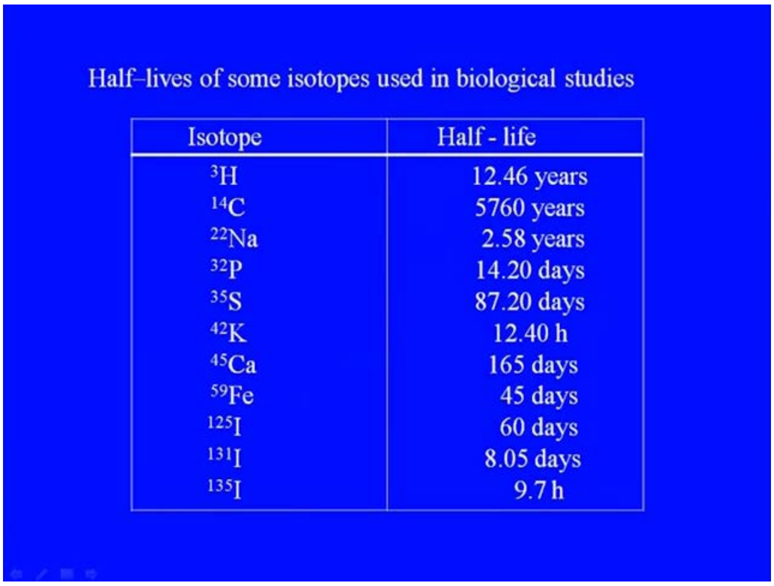
Half–lives of some isotopes used in biological studies

Isotope	Half - life
^{3}H	12.46 years
^{14}C	5760 years
^{22}Na	2.58 years
^{32}P	14.20 days
^{35}S	87.20 days
^{42}K	12.40 h
^{45}Ca	165 days
^{59}Fe	45 days
^{125}I	60 days
^{131}I	8.05 days
^{135}I	9.7 h

This table gives you half-lives of some isotopes, which are used frequently in biological study. For example, 3 H has life of almost 12.46 years. Now, 14 C has a very long half-life of 5,760 years approximately Na 22, which has a half-life of 2.58 years 32 P is 14.2 days. So, this is very much suitable for many biological works like in field of molecular biology.

Now, 35 S has 87.2 days of half-life, 42 K has 12.40 hours of half-life and likewise you can go on, like I 131 has 8 days of half-life, I 135 has 9.7 hours of half-life. Now, this is very significant, because you would when you are utilizing these radioisotopes for different application, like say in biological research or biotechnology research then if it is a too longer half-life.

It is very difficult in storage and discarding this and also utilization, but if you have a half-life like 32 P 14 days, it is becomes very important. It becomes quite good for being used in biological research, because you can after certain period the, this particular will decay its many half-lives, actually. So, this is very important half-life is a very important phenomenon in a radioisotope technique.

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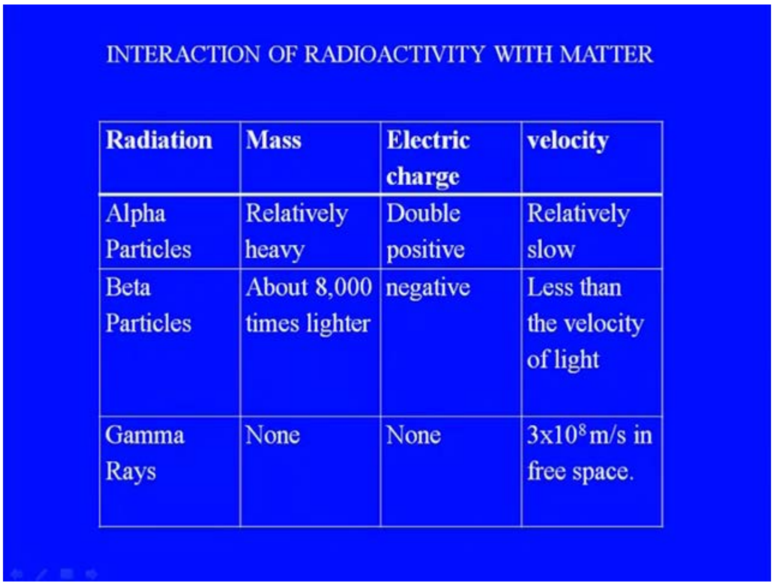

INTERACTION OF RADIOACTIVITY WITH MATTER

Radiation	Mass	Electric charge	velocity
Alpha Particles	Relatively heavy	Double positive	Relatively slow
Beta Particles	About 8,000 times lighter	negative	Less than the velocity of light
Gamma Rays	None	None	$3x10^8$ m/s in free space.

Now, let us move on to how does this radioactivity interact with matter? That is a very important part, because that is hoe detection will be done and that is how you will understand that why certain radioisotopes certain emissions like certain radioisotopes for particular emissions are not used and some are used. Like I said, alpha particle emitters are not some much used and beta emitters are frequently used.

So, let us go one by one, now alpha particles they are relatively having if you remember if you can recall they were like doubly positively charged with atomic number 2 and mass number 4. So, there are there would be certainly relatively slow. Beta particles they are lighter, they are much liter they can be negatively or positively charged and they would be faster than the alpha particle. Like way gamma rays, it does not carry any mass or charge will be highly penetrating in nature. Let us discuss each of them. How do they interact with the matter and what are the consequences in terms of the application and toxicity?

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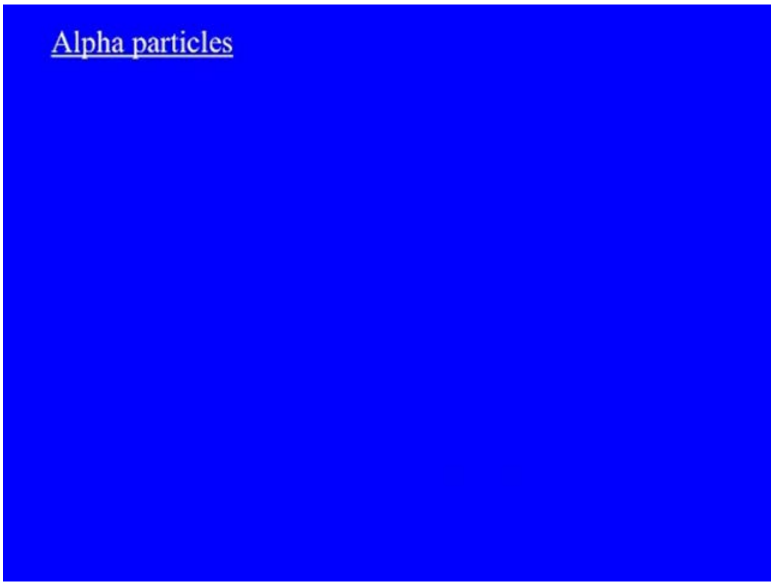

Now, alpha particles with considerable energy with all particles of a particular isotopes having same amount of energy, these particles interact with matter by causing either excitation or ionization. Now, excitation means, energy is transferred from alpha particle to orbital electrons of neighboring atom and the electrons been elevated to higher orbital and then when they come back to the original orbital then radiation will be emitted.

Now, alpha particle will continue on its path with reduced energy, as it has transferred some of the energy to the electron and the excited electron falls back to original orbital emitting energy, as photons of light invisible or near visible range. In ionization, here target orbital electron is ejected completely and the atom becomes ionized. That is you get a pair, which is positively charged ion and negatively charged electron, so that is ionization so it could be either excitation or ionization.

Now, as I said, because of their size slow movement and double positive charge, they inter act strongly with matter, because as they enter the matter and they are larger in size and slow. They are bound to more with the atoms, which are coming in their way. What happens is as they interact more, they produce large number of ions per unit length of their path and there energy is therefore, will be rapidly decapitated. So, what does that mean, is they will not highly penetrate, they are very slow they are large, they do interact with matter and so they will be not so much penetrative.

For example, if you consider a five million electron volt of alpha particle, it will only travel about only 3.6 centimeter near and it not able to penetrate, even an ordinary piece of paper. So, that is how alpha particle interacts. Now, they can interact with either nuclei or orbital electron. Now, when passing in the vicinity of nucleus, it may be deflected with no change in energy, which is called Rutherford scattering or deflect with small change in energy or absorbed by nucleus, causing nuclear transformation. So, lot of thing could happen.

Now, the most probable process involved in the absorption of alpha particle, however are ionization and excitation of orbital electrons. Since, the alpha particles are low in penetration ability they are usually not hazardous for external exposure like is said, it does not enter even a piece of paper, thin paper, but if the alpha emitting nuclide is deposited in an organism then its toxic.

So, when internally deposited alpha particles are more damaging than most other types of particles, because comparatively large amounts of energies are deposited with in a very small volume of tissue. So, they will quite toxic and damaging for the tissues. So, in summary, alpha particles, they are slow they are large and they are not very penetrating dissipating their energy fast and they could be toxic in certain cases.

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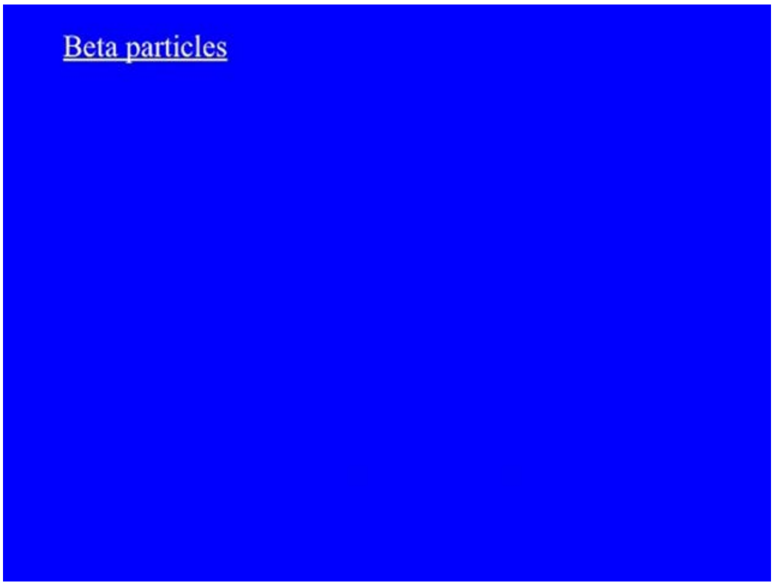

Now, let us see about how a beta particle, we are not going to discuss about positron like I said, it has a transient survival, it like annihilates very fast. But, you will be discussing about the negatron or beta particle, which is negatively charged. Now, these beta particles are small and rapidly moving particles compared to alpha particles, they are small and much lighter, they carry a single negative charge and they interact with matter by causing either excitation or ionization.

Now, beta particles can inter act with electrons as well as nuclei. Beta particles passing near nucleus will be deflected by the coulomb forces and loss of the particles, kinetic energy may or may occur coulombs repulsion between the beta particle. Electrons frequently results in ionization.

Now, in the ionization process the beta particle loose an amount of energy equal to the kinetic energy of the electron plus the energy used to free it from the atom. So, ionization will certainly, beta particles in ionization process will lose the energy. Now, so beta particles may also cause excitation of external orbital electrons, as we have seen in case of alpha particle.

So, what happens then which in turn leads to the emission of sometimes UV photons. Now, like alpha particle beta particles have a characteristic, average travelling distance through matter and that is dependent upon their initial kinetic energy. Now, beta particle with energy about two million electron volt will travel up to almost 9 meter in air and about 10 millimeter in water. So, comparatively beta particles will have higher penetrating power as they are light and before decapitating there energy, they move to rather more distance as compared to alpha particles.

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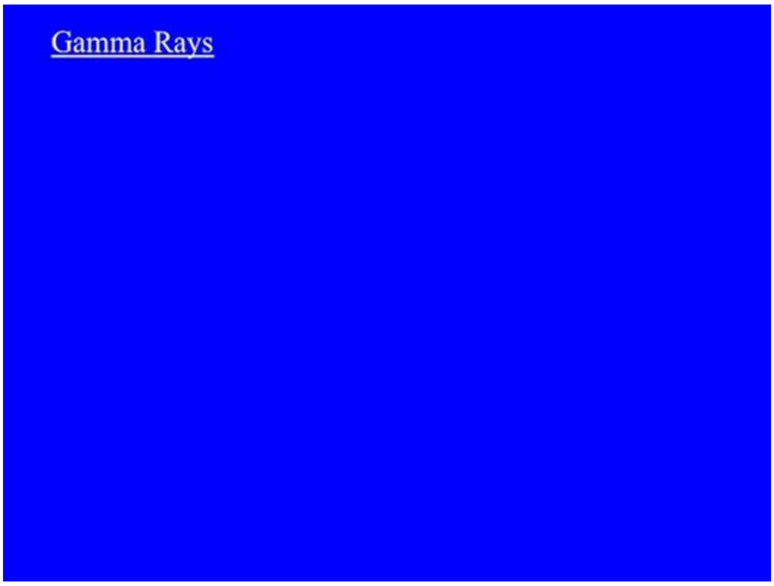

Now, gamma rays. Let us come to third one, that is electromagnetic radiation, which is gamma rays. Now, these are electromagnetic radiation like I said and have no charge or mass, they rarely collide with neighboring atoms and they are highly penetrating. Now, their interaction of photons with matter involves several distinct process and we will discuss each of them one by one.

Now, relative importance and efficiency of each process is strongly dependent upon the energy of the photons and they are like, we will see they are low energy, medium energy and high energy gamma photons. Also, it will depend upon the density and the atomic number of the absorbing medium. Now, three of the methods which lead to the production of secondary electrons, which in turn can cause excitation and ionization. So, gamma rays by itself is not ionizing or involved in excitation, but through electrons secondary electrons will be generated, as we will discuss, it is involved in excitation and ionization.

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Now, there is one called Rayleigh Scattering. In Rayleigh scattering, when a photo interacts with atom it may or may not impart some energy to it. The photo may be deflected with no energy transfer that is elastic interaction and this process is called Rayleigh scattering.

So, this not that important here, but the other three which we are going to discuss will show you, how they will interact and cause different phenomenon. One is photo electric absorption, in photo electric absorption low energy gamma rays will interact with orbital electron. Now, in this process the photo transfers all of its energy to the electron and its own existence terminates.

So, the electron will escape it s orbit with a kinetic energy equal to the difference between the photon energy and its own binding energy and will behave like negatron. As we have discussed, it will behave and do all this as negatron. So, what is happening here, that there is a low energy gamma ray interact with electron and will transfer its complete energy and it particular existence will not be there anymore and that orbital electron will be ejected and behave as negatron.

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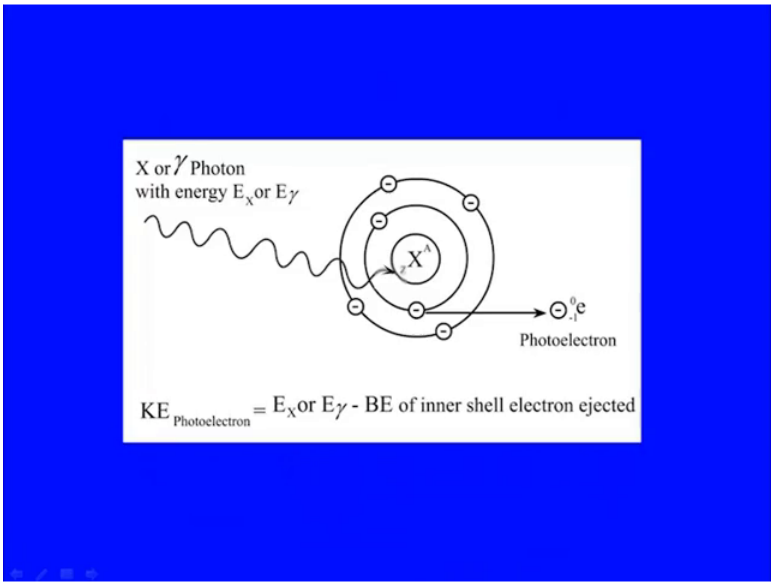

Now, let us see how does it this figure shows that particular phenomenon. Here, that there is a gamma ray coming, it will eject this electron from here, transfer its whole energy and the electron is ejected as a photo electron, in this case. So, low energy gamma ray will have a particular way of interacting. Now, let us see the medium energy gamma ray.

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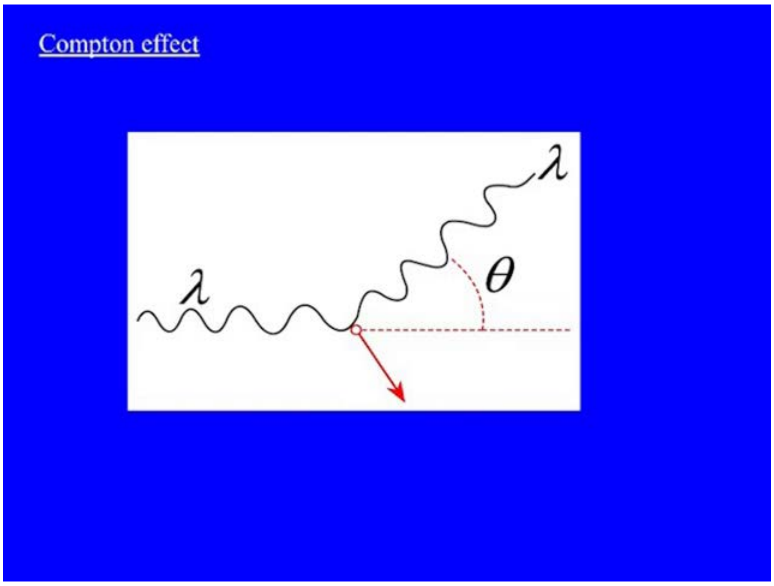

Here, it is this particular effect is called Compton effect. In this process the photon interacts with an atomic electron sufficiently to eject it from orbit. The photon retains a portion of its original energy, remember this is a medium energy and not the low energy photo, and it will continuously it will move continuously in a new direction because it will be deflected, actually.

Thus the Compton effect have a absorption component as well as scattering component the amount of energy lost by the photon, can be related to the angle at which the scattered photo travels relative to the original direction of travel. So, here in the Compton effect you have energy partly transferred for ejection of electron or negatron and partly some energy carried or it is carried over as a low energy gamma ray.

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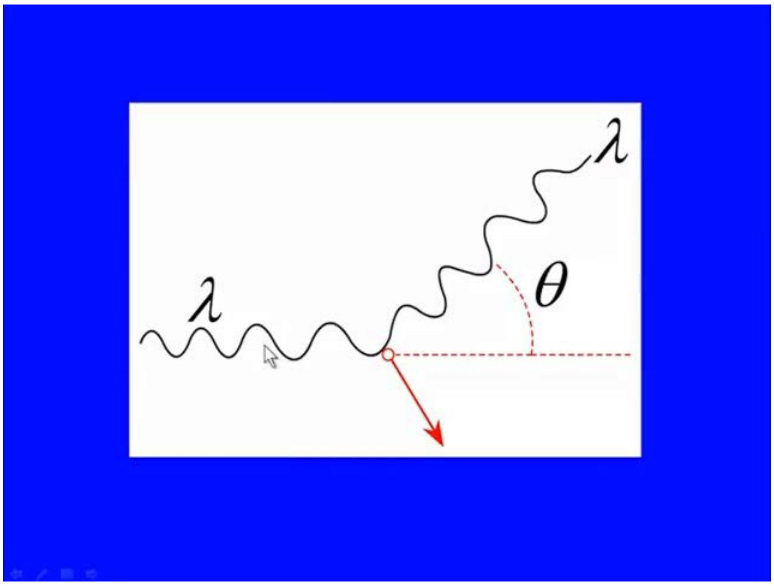

Let see try to understand this in figure. So, what happens that you have a gamma photon, with medium energy it hits orbital electron, electron is ejected and the gamma energy ray continues in a new direction here. This again can act as a low energy photon.

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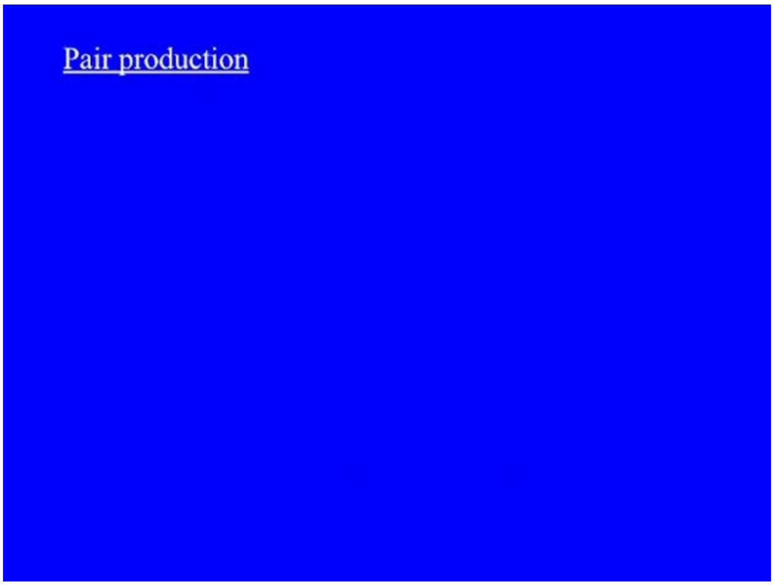

Here, now third one is Pair production. Now, this Pair production occurs when there is high energy gamma rays. So, what happens high energy gamma rays interact with nucleus of an atom and all the energy of the gamma ray is converted to positron and negatron. So, remember here rather than ejecting a simple electron orbital electron, here it is interacting with nucleus of an atom and its all energy will be converted to positron and negatron.

Now, photons with energy greater than 1.024 million electron volt under the influence of the electromagnetic field of a nucleus may be converted into electron and positron. Now, at least this much of energy of photons are required for a pair production and because it is equaling to point 0.51 million electron volt each and then pair production will occur.

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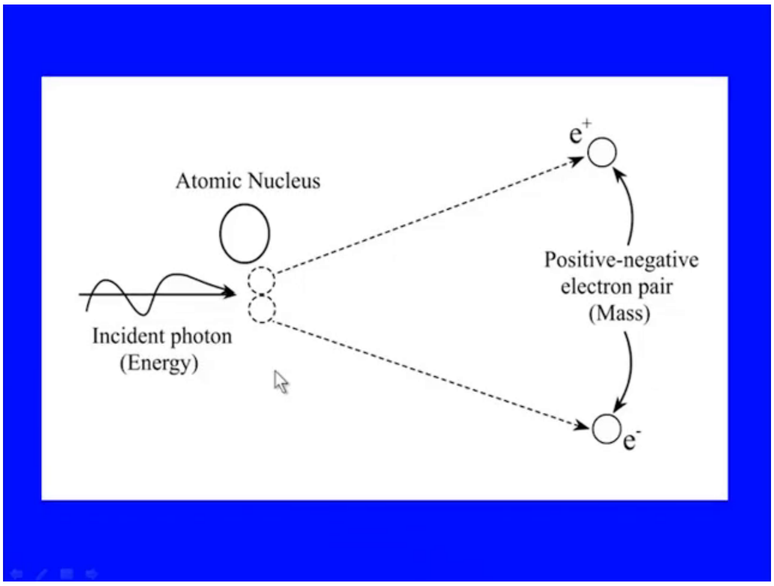

If you can see here, in this figure there is an incident photo, which is high energy photon influence of atomic nuclei, it is converted to positively charged and a negatively charged particles, which is negatron and positron. So, this only happens with the only happens with the high energy gamma rays. Also, there are whole lots of different thing which will interacting.

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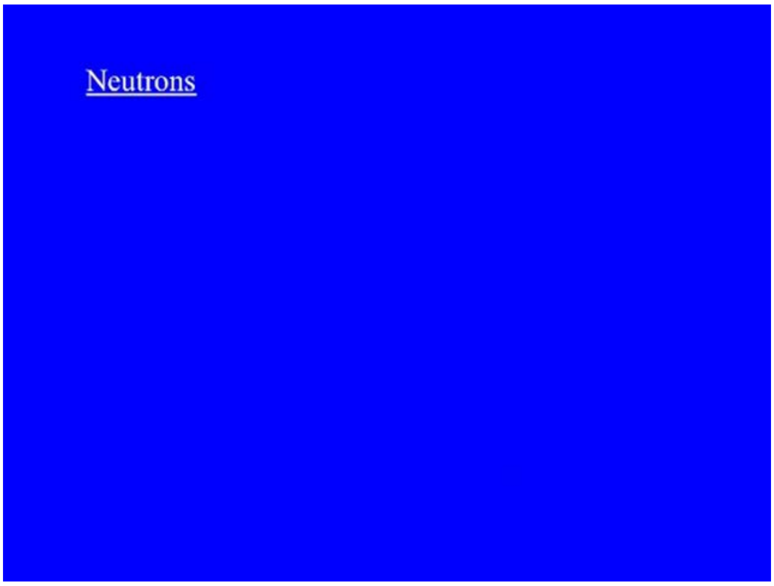

Neutrons interact and encounter biological material it is utilized in for lot of biological applications and it will collide with a proton with a sufficient force to disloss the proton from the molecule. A proton may then have sufficient energy to travel some distance and the tissue causing secondary damage through ionization and excitation. We are not going into detail of that, but this is also an important particle which could be utilized or which can inter act with matter and it is utilized in like say neutron diffraction and lot of other techniques.

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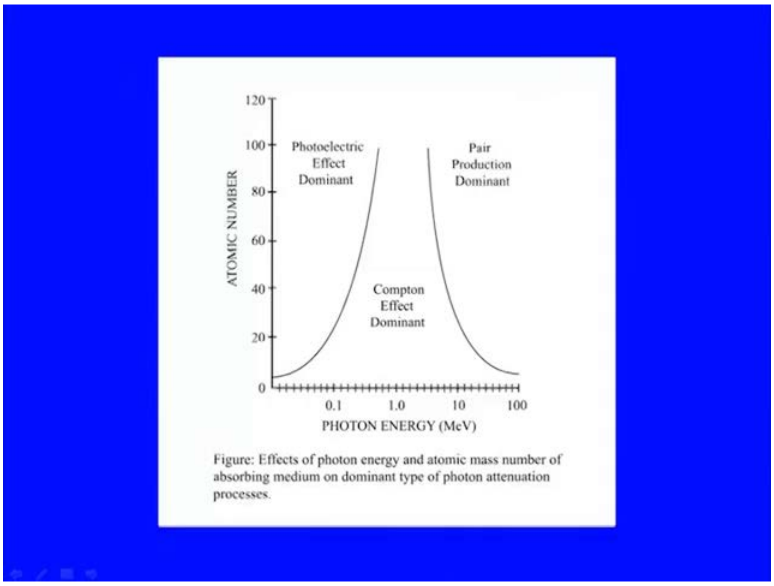

This figure here, summarizes the inter action of gamma rays with the matter. Now, if you can see here, this is like atomic number and the x-axis is the proton energy. If you can see it, in different areas are distributed in here, so there is a low energy area which had dominant photo electric effect that what is that means that low energy gamma rays will show photo electric effect, the medium energy gamma rays will show Compton effect dominantly and the high energy gamma rays will show the pair production here. So, you have three kinds of effects of gamma radiation, which are shown in here and summarizes the whole interaction very well, all right?

So, in this lecture what we have summarized is many different basic concepts. One is that how in terms of energy the radioactive decays are measured, which we have said it is in terms of electron volt or which is million or mega electron volt, which is larger unit, because electron volt is a very small unit. We have seen, what are the units of radioactivity, which is expressed in terms of Bacquerel, which is giga bacquerel or terra bacquerel, and also it, is widely used in terms of Curie, where you use micro Curie or mille Curie.

We have seen that the energy of, we have seen about beta spectrum in alpha and gamma there is a range of energy of radioactive decay. But, the beta emitter, they emit radiation in a wide range from zero to very to a maximum value. For each of a typical radioisotope of beta emitter we have seen the interaction of the different radioactivity with matter. As we have seen that you have alpha particle, which is a heavier particle, which is doubly positively charged and slow, it interacts rapidly, it interacts quite a lot with the matter as it enters the matter. And will dissipate energy very fast and will cause ionization or excitation.

Like I said, it is quite toxic if ingested, because it will cause too much fat in a very small area and can have can create tissue damage and other problems. The beta particles are slow entities and they are faster than the alpha particle and they are the ones, which are used most in biological world of different energies, like it could be low energy beta particle emitter or a medium energy beta emitter or high-energy beta emitters.

We will discuss them as we go along then there is electromagnetic radiation gamma radiation, which in itself does not carry any mass or charge but it also cause ionization or excitation through the beta particles, which is the production of which takes place through various effects, which we have discussed like photoelectric effect, compton effect or pair production. So, this completes our here the basic concepts, which we were discussing.

Now, we will move onto the particular methods of detection and quantification, in next lecture. Now, there are methods, which we are going to discuss one by one. One based on the gas ionization, where the g m counters have been developed to detect the radio activity and we will see what are different types of these counters and how they are useful, then we will be discussing about the methods based on excitation. So, one is ionization like if you could recall we have told you both beta particles or alpha particles and gamma radiation can cause both ionization and excitation on.

Methods based on excitation will be discussed where scintillation counters have been developed and we will be discussing them in detail then another. One-third important method, autoradiography that is exposure to of radioactivity to photographic film that we are going to discuss in subsequent lectures, so this we complete here.

Thank you.