Host-Pathogen Interaction (Immunology)
Prof. Himanshu Kumar
Department of Biological Sciences
Indian Institute of Science Education and Research, Bhopal

Module No # 08
Lecture No # 38
Pattern – Recognition Receptors – RLR Signaling Pathways

Hi, in previous session we have discussed about the RLR discovery of RLR members particularly RIG-I. And we discussed about the endogenous ligand as well as exogenous ligand which is mainly derived from different viruses. And this exogenous infection means the viral~~bration~~ infection also result to the disturbance of RNA metabolism and that result to the activation of RLR signaling pathway.

So in this session we will discuss about the RLR signaling pathway which is I will show you or your understanding I will make it extremely simple. However this pathway is extremely complicated not only this pathway the previous TLR and whatever pathway you study. I will just touch up on the complexity I will just show you couple of slides in order to understand the complexity this RLR and signaling pathway as well as the complexity of activation of these members, RIG-I and MDA5~~FI~~.

And downstream there is also lot of complicated I will not show you those complication here I will just show you the complication in activation or deactivation of RIG-I and MDA~~F~~5. The more complicated pathway is basically not needed to you it is very much needed to the researcher who are working in this field. So let us begin with this RLR signaling pathway.

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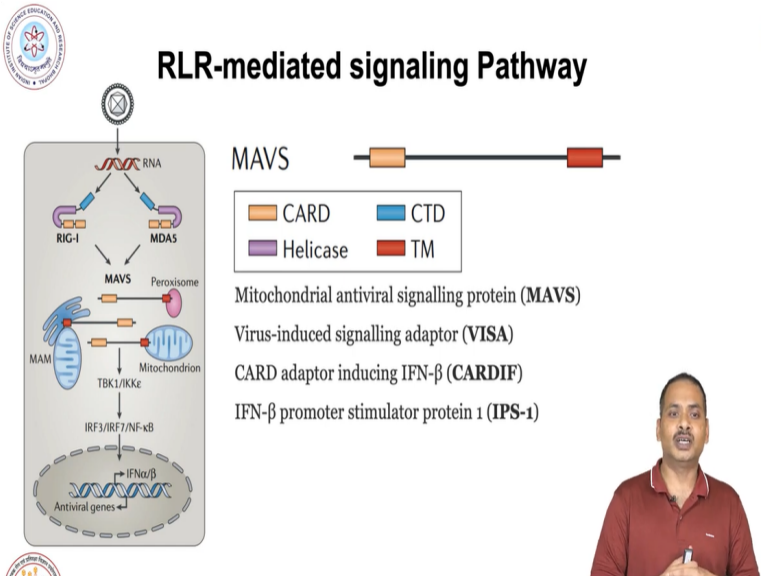

So this RLR signaling pathway is basically activated when there is a virus infection. So once this virus is infecting the cell this RNA is transported into the cell in initially it will be in endosomes or phagofragolysoizosomes and then this RNA will be also reacich in the cytoplasm. Over there this RNA species will be sensed by either RIG - I or MDFA5. So this is a simple schematic of RLR signaling pathway here you can see that the RIG - I and MDFA5 which is initially present in inactive form.

As I told you in previous session that this CTD this CTD domain carboxyl terminal domain basically this whole molecule is present in close format. Close format means neither this card will be exposed nor this helicase will be exposed when there is no infection. But when this infection comes then this there will be conformational changes in these molecules will open up in order to sense the ligand and also to activate the downstream signaling.

So this signaling is initiated to RIG-I and MDAF5 and then there will be recruitment of adapter molecule. For all signaling model molecule there is a one simple scheme there will be s receptor molecule there will be a adaptor molecule and there will be some kinase. And then there will be activation of transcription factor that eventually result to the effector response. This is a common feature of almost all signaling pathway so here the adopter molecule is MAVSmass and this massMAVS is also known as a there are various name of MAVSmass.

So you will understand that time when this RLR pathway was dissected out there are so many researcher all over the world they are working in order to discover this signaling pathway. So that is why molecule massMAVS or the adopter molecule of this RIG-I and MDFA5 pathway

this as a four name I will show you in a short while. So this adopter molecule is basically localized over here you can see this is localized on peroxisomes.

And it is present on mitochondrial outer membrane and it is also present in MAM this is mitochondrial associated endoplasmic reticulum membrane. Some people use this word endoplasmic reticulum or ER or some people do not use. So it is in a very simpler form the membranes which are associated with the mitochondria over there this adopter molecule is present.

So since it is located on the mitochondria people previously they thought that it is playing important role in apoptosis. Probably you might be aware that this process of apoptosis uses the mitochondria or the some molecules which is present in mitochondria. So of course a lot of extensive research was performed and then people do say that this molecule do have some role in apoptosis. Anyway, we are not here to discuss the apoptosis let us go to the signaling pathway.

So once this molecule the RIG-I and MDF5 MDA5 is activated this activated molecule is recruiting this adapter molecule. And here you will see that the RIG-I and MDF5 they have a card and this adapter molecule also have a card. And this card is basically interacts with the RIG-I and MDF5 card and this basically a homotypic interaction. And once this interact that this signalosome (()) (07:41) the is complex protein activate both enough NF- Kappa B as well as this will also activate the IRF interferon regulatory factors.

The interferon regulator factor is IRF3 or and IRF7 as you can see and the activation of this NF- Kappa B is this signalosome is home basically activate towards end it will activate the IKK complex. As you have seen in previous session in TLR signaling this IKK complex is consist of 3 members IKKA alpha, beta and gamma. And then once this get this complex get activated this will phosphorylate get activated this will phosphorylate I Kappa B and this I Kappa B will undergo proteosomal mediated degradation after ubiquitination.

And a free N enough Kappa B will be translocated into the nucleus and then there will be a transcription of inflammatory cytokine. On another hand this if you remember the TRIF drift mediated signaling so this activates the kinase TBK1 or IKKI or IKK epsilon. So once this kinase gets activated this kinase will phosphorylate interferon regulatory factor 3 or 7. And

then once it is phosphorylated then this IRF will make a dimer and translocate into the nucleus and then there will be transcription of type 1 interferons.

So in that way the signaling pathway activates so this adopter molecule you can see them MAVS~~mass~~ again they have this card domain. The card this has a card as well as trans membrane domain that is why this is localized on peroxisome mitochondria or outer membrane of mitochondria for membranes associated with mitochondria. So this ad~~a~~opter got several names because of discovery this adopter was discovered by 4 independent groups in the world and they gave their own name.

So that is why this molecule has a 4 name one is MAVS~~mass~~ this is mitochondrial antiviral signaling protein. Another is VISA this virus inducing signaling adaptor and another name is this CARDIF which is stand for card adapta~~o~~r inducing interferon beta. And there is a molecule finally the other name is IPS-1 this is interferon beta promoter stimulator protein 1. So this molecule got several name and we also discovered this molecule when I was doing research in Japan.

So we discovered these molecules ~~inter~~ IPS-1 this adaptor molecule and we gave this name IPS-1. So this is the simple presentation of a RLR signaling pathway here I just give you the glimpse of complexity.

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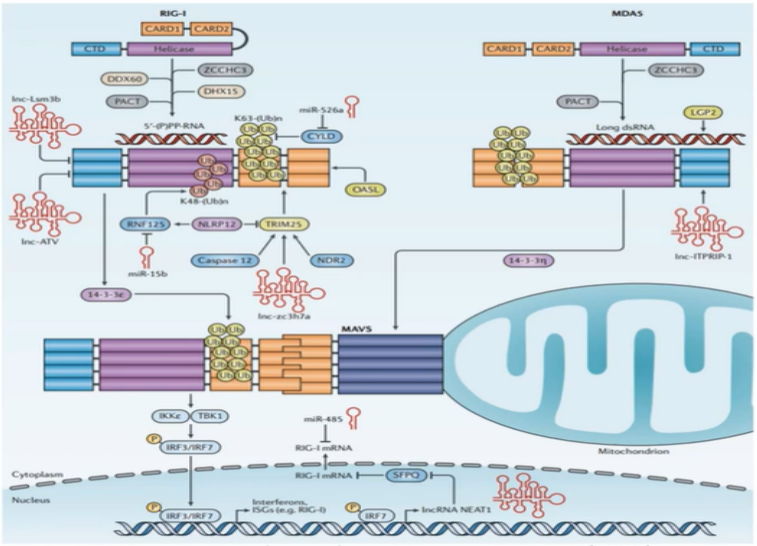

Nature Reviews Immunology **volume 20**, pages537–551 (2020)

So here you can see how complex this signaling pathway is? There is a RIG-I and this RIG-I get oligomerize upon activation RIG-I or MDA~~-~~5 and then there will be recruitment of adaptor molecule known as IPS1. And this is regulated by plethora of molecule there are so

many molecules and these molecules are protein and this protein as a regulator and there are some long non-coding RNA which is also regulating this pathway.

At the bottom you can see there is one more molecule which is regulating the transcript of RIG-I that is there is a micro RNA. If you see carefully at the bottom then you will find out that there is a molecule known as micro RNA 485 which regulate the expression of RIG-I. So we have discovered this molecule this micro RNA 485 and this molecule plays a very important role in regulating the RIG-I as well as this molecule the micro RNA 485 very interestingly we found that this also control the replication of influenza virus.

And how it is controlling? Basically, they directly binding to the transcript which is a essential for making polymerase and this polymerase is needed in order to make the copies of influenza virus. So this micro RNA binds with those transcripts and since it is binding there will be a reduction in expression of those transcripts and in that way the viral replication is very strongly checked by this micro RNA 485.

So this is the one snapshot of signaling and if you see this RIG-I and MDA5 this both molecules are very tightly regulated.

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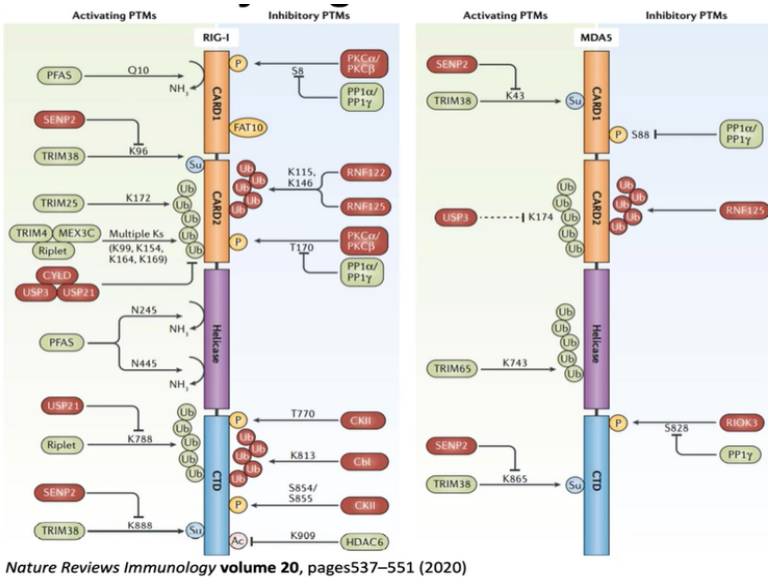

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As you can see in this slide there are so many post translation modification which activates the RIG-I there are so many post translation modification which inactivate these members that is RIG-I and MDA5. So overall what I want to say that this is the extremely complex signaling pathway and in your current scenario you need not to worry about complex signaling pathway.

And if you interested and if you want to pursue research in this field then of course you need to understand this complexity with this I am stopping here and in next session I will discuss about the RLR and disease and I will also discuss about Therapeutics thank you.