The Micro Fugitive: Understanding Herpesvirus Targets that impact Viral Latency

Francesca Martin

Herpesviruses are notoriously good at hiding, so much so, that people live many years without knowing they have an infection. What makes these viruses so efficient is their ability to remain latent within the body, only surfacing occasionally to spread via shedding (think cold sores or skin lesions). Their name itself derives from the greek word '*hérpein*' which means 'to creep' and that's essentially what they do, slowly spreading from cell to cell with little detection from the immune system. Their ability to manipulate and amend their environment to their own benefit has proven useful as they often establish a life-long infection. Due to their ability to persist, it's important to try and understand how they work on a molecular level. Should this be possible, potential targets can be identified and therapies produced.

This project focusses on human herpes virus 8 (KSHV) and how it manages to hide within human tissue without being detected by the immune system. We know that it produces a protein called LANA during infection and this plays a large role in maintaining the presence of the virus. We combined the genomes of two viruses to create a new virus that could infect mice (MuHV-4) but produce human proteins (LANA) that are important in establishing continuous infection otherwise known as latency. Deletions were made in the genome of our mutant viruses in an area critical for latency and we did this in order to investigate as to how these deletions affect viral latent behaviour. After infecting mice with these viruses, their spleens were then taken out after 14 days and analysed to see how much of the virus had been reactivated. Currently, there are few efficient models capable of permitting the study of KSHV. These deletions were made in an area that codes for the LANA protein and are important for latency. The deletions also differed in size to understand how important this region is and cover an area that is critical in DNA replication and episomal persistence.

The episome is a small extra piece of DNA separate from original viral DNA that can replicate independently. These episomes are important since they are good at integrating themselves into the host and maintain the viral infection even though the initial infection may have been cleared by the immune system. These deletions are vital for the episome to replicate and therefore it's important to understand this better. The LANA protein itself is 1162 amino acids long and deletions were made between amino acids 262 and 320. It has been reported previously that making deletions between 262 to 320 can reduce viral latency in cells, however we wanted to take it one step further and see how these viruses behave in live animal infection. It's important to understand what drives viral latency but, more so, what areas of the viral genome are critical to viral persistence. In targeting a sequence that is critical in latency, any deletions should theoretically impede viral ability to establish prolonged infection. This study discovered that these these mutant viruses were still able to establish an infection in both a live animal model and in cultured cells. However, deletions did not impede the rate at which viruses were growing nor were there any significant growth differences found between each mutant. This suggests that the assays implemented may not be sensitive enough to detect such slight deletions or it may be possible that the LANA protein is compensating for those deletions in other ways not yet known.

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Biology Education Centre, Uppsala and Instituto de Medicina Molecular, Lisbon
Supervisor: Dr. Pedro Simas