Introduction

Human cytomegalovirus (HCMV) is a widespread pathogen that can lay dormant and rarely causes disease in healthy individuals. However, in immune-compromised individuals, such as transplant recipients and AIDS patients, HCMV infection can be life-threatening.

In this study, we examined one viral gene, US27, which encodes a protein that is similar to human chemokine receptors. Chemokine receptors have seven transmembrane domains and signal immune cells to sites of infection. Previous work in our lab found that US27 binds to GABARAP (γ-aminobutyric acid receptor-associated protein), a 14kD cellular protein that plays a role in receptor trafficking.

We used fluorescence microscopy to examine the US27-GABARAP interaction in human embryonic kidney cells (HEK293). GABARAP has been shown to bind to a WXXL motif in target proteins, and this sequence is present in the C-terminal domain of US27. Using a series of proteins with mutations in this motif (Figure 1), we found that the WXXL motif in US27 is required for both GABARAP binding and for localization to the endosomes.

Results

US27 co-localizes with GABARAP; US27 mutants do not

US27 localizes to the endosomes; US27 mutants do not

Fluorescence microscopy showed that the interaction of US27 and GABARAP requires a WXXL motif found in the C-terminal domain of US27.

Mutation of the tryptophan (position 306) and leucine (position 309) residues in the C-tail of US27 disrupts its binding to GABARAP and localization to the endosomes. The interaction of wild-type US27 with GABARAP is depicted in Figure 4.

These results show that US27 functions primarily as an intracellular protein. Our findings suggest that the viral receptor could induce internalization of cellular chemokine receptors and may modulate host chemokine signaling responses.

Future work includes identifying additional proteins that are part of the US27-GABARAP complex with the aim of clarifying the role of US27 during virus infection.

Conclusions

Fluorescence microscopy showed that the interaction of US27 and GABARAP requires a WXXL motif found in the C-terminal domain of US27.

Mutation of the tryptophan (position 306) and leucine (position 309) residues in the C-tail of US27 disrupts its binding to GABARAP and localization to the endosomes. The interaction of wild-type US27 with GABARAP is depicted in Figure 4.

These results show that US27 functions primarily as an intracellular protein. Our findings suggest that the viral receptor could induce internalization of cellular chemokine receptors and may modulate host chemokine signaling responses.

Future work includes identifying additional proteins that are part of the US27-GABARAP complex with the aim of clarifying the role of US27 during virus infection.

Acknowledgements

This work was supported by funding from the National Institutes of Health and USF Faculty Development Funds (to Juliet V. Spencer).