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Translational control of APOBEC3G/F Restriction Factors by the HIV-1 Vif protein

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BACKGROUND

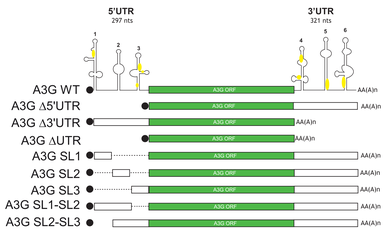
The human immunodeficiency virus type 1 (HIV-1) requires the concerted contribution of many cellular factors to achieve efficient replication. Similarly, mammalian cells express a set of proteins called restriction factors to suppress viral replication. Among these factors, the family of APOBEC3 (Apolipoprotein B mRNA editing enzyme catalytic polypeptide-like 3 or A3) proteins and in particular A3G and A3F are the most efficient against HIV-1. They belong to a large family of cytidine deaminases that catalyze the deamination of cytidines to uridines in single stranded DNA substrate during HIV-1 retrotranscription and is lethal for the virus. The antiviral activity of A3G/F is counteracted by HIV-1 Vif (Viral infectivity factor) protein. Vif significantly reduces their expression in cell and their incorporation into viral particles by 1) recruiting an E3 ubiquitin ligase complex to induce their degradation by the proteasome, and 2) regulating their translation. Up to now, the mechanisms by which Vif regulates the translation of A3G/F are not known.

METHODS

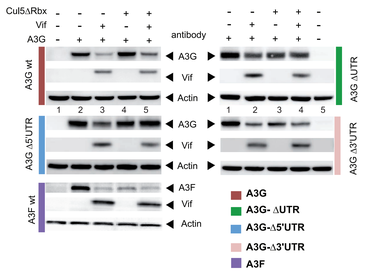
To address the role of Vif in the regulation of A3G/F translation, we tested the importance of the unstranslated regions (UTRs) of A3G/F mRNA in the translational inhibition. HEK 293T cells were transfected with wild-type and mutated constructions of A3G and A3F mRNAs (ΔUTRs, Δ5'UTR, Δ3'UTR and ΔSL) in presence or absence of Vif. These experiments were also performed with proteasome inhibitors (Cul5Δ Rbx, ALLN) in order to distinguish the proteasomal degradation pathway from the translational inhibition.

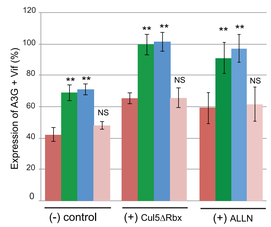
RESULTS

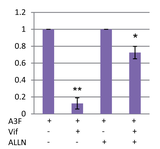
Secondary structure of A3G mRNA and constructs



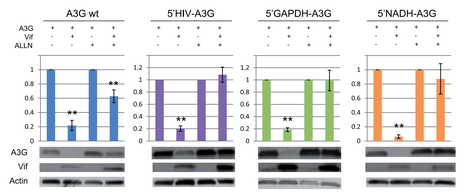
The 5'UTR of A3G and A3F is essential for the translational regulation



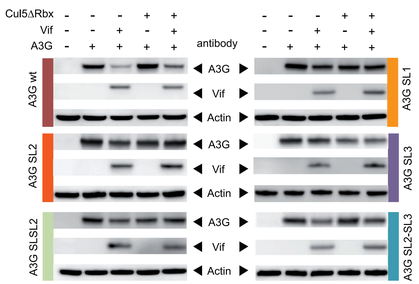


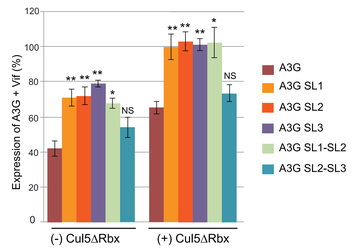


The translational inhibition is specific of the 5'UTR of A3G

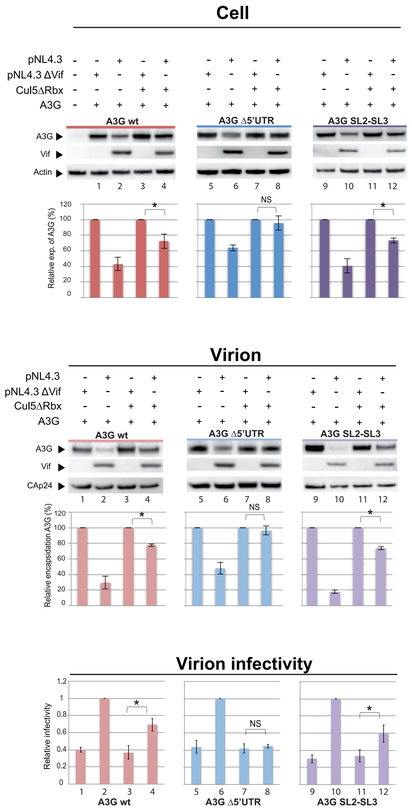


SL2 and SL3 are important for the translation inhibition

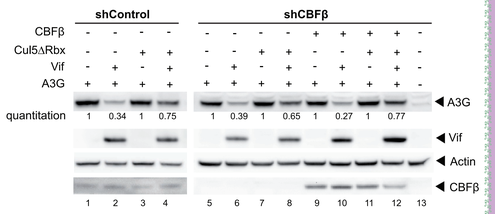




The translational inhibition restores HIV-1 infectivity



The translational regulation is independent of CBF-β



1- Expression of A3G and A3F in HEK 293T cells, with or without proteasome inhibitors, shows that the 5'UTR of A3G/F mRNA is crucial for the Vif-induced translational regulation.

2- Within the 5'UTR of A3G mRNA, stem-loops 2 and 3 are required for the translational inhibition of A3G translation by Vif.

3- The Vif-induced translational inhibition of A3G reduces the packaging of A3G and partially restores viral infectivity.

4- The transcription factor CBF-β (Core Binding Factor β), which is known to stabilize the Vif/A3G complex, is not essential to induce the translational inhibition of A3G by Vif.

CONCLUSION

This study shows a second mechanism by wich Vif decreases A3G/F expression in cells. This translational inhibition is an A3G 5'UTR-dependent mechanism and can increase HIV-1 viral infectivity. Thus, preventing the translation inhibition mechanism of A3G/F may restore the cellular restriction activity of A3G/F.

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Dr. Reuben Harris (University of Minnesota, Minneapolis, MN-USA) for the HEK293T stable cell lines (shCBFb and shControl).

Reference:

- Mercenne G. *et al.*, *Nucleic Acids Res.* 2010 *38, 633-646 HIV-1 Vif binds to APOBEC3G mRNA and inhibits its translation.*

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