Elucidating the formation of a multimeric protein complex with HTLV-1 Tax and its impact on viral replication

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Background:
The human T-cell leukemia virus type 1 (HTLV-1)-encoded transactivator Tax-1 is essential for HTLV-1 replication. We recently found that Tax-1 interacts with transcription elongation factor ELL2 in the nucleus to enhance viral transactivation.

Methods:
Here, we aimed to characterize Tax-1:ELL2 complex formation and its impact on viral transactivation by bioinformatics, confocal imaging, co-immunoprecipitation (co-IP), and luciferase assays.

Results:
We found that Tax-1 and ELL2 not only co-precipitate, but also co-localize in dot-like structures in the nucleus. Co-IP revealed that Tax-1 deletion mutants lacking either N-terminal (aa 1–37) or C-terminal regions (aa 150–353) of Tax-1 were impaired in interacting with ELL2. Contrary to Tax-1, the related, non-oncogenic Tax-2B from HTLV-2B did not interact with ELL2. ELL2-R1 (aa 1–353), which carries an RNA polymerase II binding domain, and ELL2-R3 (aa 515–640) were sufficient to interact with Tax-1; however, only ELL2-truncations expressing R1 could enhance Tax-1-mediated transactivation of the HTLV-1 promoter. The closely related HIV-1 regulatory protein Tat recruits ELL2 and the super elongation complex (SEC), which partly consists of the positive transcription elongation factor b (pTEFb; CDK+cyclinT1), to reduce promoter proximal pausing through phosphorylation of RNA polymerase II. Here, characterization of the multimeric Tax:ELL2 complex revealed that Tax interacts with CDK9, cyclin T1, and both proteins, confirming earlier observations. Moreover, we identified a new Tax:ELL2:cyclin T1 complex, with both endogenous cyclin T1 and transfected cyclin T1. Interestingly, the co-IP of Tax with CDK9 was shown to decrease in the presence of cyclin T1, suggesting a competition between cyclin T1 and CDK9 for Tax binding.

Conclusion:
Taken together, this study identifies domains in Tax-1 and ELL2 being required for Tax-1:ELL2 complex formation and for viral transactivation and the current data strongly suggest the existence of a SEC-like complex in HTLV-1, but its detailed composition remains to be determined.

Disclosure of Interest Statement:
Nothing to declare.