Breaking the Genome

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Human papillomaviruses (HPV) are oncogenic DNA viruses that include the causative agent of cervical cancer (α-HPV), and some viruses that are associated with non-melanoma skin cancer (β-HPV). α-HPVs must persist in cells to cause tumors; however, β¬-HPV persistence does not appear to be necessary for tumor maintenance. This suggests that β¬-HPV may act as a co-factor to increase the oncogenic potential of ultraviolet light in skin cells. In a recent study published in Virology, Drs. Nicholas A. Wallace and Denise A. Galloway (Human Biology Division), in collaboration with researchers from the University of New Orleans and Tulane University, demonstrate that downregulation of ATM by the β-HPV protein E6 inhibits Long INterspersed Element-1 (LINE-1) retrotransposition, but does not inhibit double strand breaks in DNA generated by LINE-1, suggesting a potential mechanism for mutagenesis in β-HPV infected cells.

LINE-1s are a family of retrotransposons occurring in large numbers in eukaryotic genomes. LINE-1s encode two genes that enable replication and dispersal through the genome by a “copy and paste” mechanism. One gene encodes a reverse transcriptase that generates a DNA copy of LINE-1 RNA, which is then reinserted into the genome through double strand breaks (DSBs) generated by another gene product, the LINE-1 endonuclease (see figure).

The HPV protein E6 is one of two primary oncogenes in high-risk α-HPV; among other functions E6 disrupts DNA damage repair by targeting p53 for proteasomal degradation. In contrast, β-HPV E6 indirectly disrupts p53-mediated DNA damage repair by reducing expression of the p53 modifying enzymes ATR and p300 (Howie, et al., 2012 and Wallace, et al., 2012). Since ATR is a member of the PI3 kinase family, the authors first sought to determine whether β-HPV E6 also downregulated the expression of the related PI3 kinase ATM. Transduction of β-HPV E6 protein significantly decreased ATM levels. However, both an E6 mutant unable to bind p300 or siRNA knockdown of p300 did not reduce ATM expression, suggesting that the reduction of ATM in these cells is mediated through a direct interaction of β-HPV E6 with p300, similar to the previously reported mechanism for ATR reduction by β-HPV E6.

Previous studies suggested that ATM is involved in LINE-1 retrotransposition, although it was unclear whether or not this is a direct effect. Wallace et al. found that PI3 kinase inhibitors that

specifically block ATM activity dramatically reduced LINE-1 retrotransposition. Supporting this conclusion, the team found that transfecting an ATM kinase dead mutant reduced LINE-1 retrotransposition by 50%. The authors next sought to characterize the mechanism for this decrease in retrotransposition. They found that ATM deficient cells displayed significantly less LINE-1 mediated cytotoxicity, a phenotype also observed in cells expressing β-HPV E6. One possible explanation for this reduced toxicity may be that reduced ATM levels decrease the frequency of DSBs; however, the authors found no difference in the number of DSBs induced by LINE-1 endonuclease in ATM-positive or ATM-negative cells.

Taken together, these data prove that β-HPV protein E6 reduces ATM expression levels and, as a consequence, reduces LINE-1 retrotransposition in the genome. However, LINE-1s can act as an endogenous mutagen by generating DSBs in the genome, and ATM depletion does not inhibit this activity. “In this work, along with our collaborators, we were able to show that β-HPVs can disrupt the repair of damaged DNA in a way that may enhance the deleterious effects of LINE-1 expression,” said Dr. Wallace.

Wallace NA, Gasior SL, Faber ZJ, Howie HL, Deininger PL, Galloway DA. 2013. HPV 5 and 8 E6 expression reduces ATM protein levels and attenuates LINE-1 retrotransposition. Virology. Epub before print, doi: 10.1016/j.virol.2013.04.022.

See also: Howie HL, Koop JI, Weese J, Robinson K, Wipf G, Kim L, Galloway DA. 2011. Beta-HPV 5 and 8 E6 promote p300 degradation by blocking AKT/p300 association. PLoS Pathog. 2011 Aug;7(8).

See also: Wallace NA, Robinson K, Howie HL, Galloway DA. 2012. HPV 5 and 8 E6 abrogate ATR activity resulting in increased persistence of UVB induced DNA damage. PLoS Pathog. 8(7).

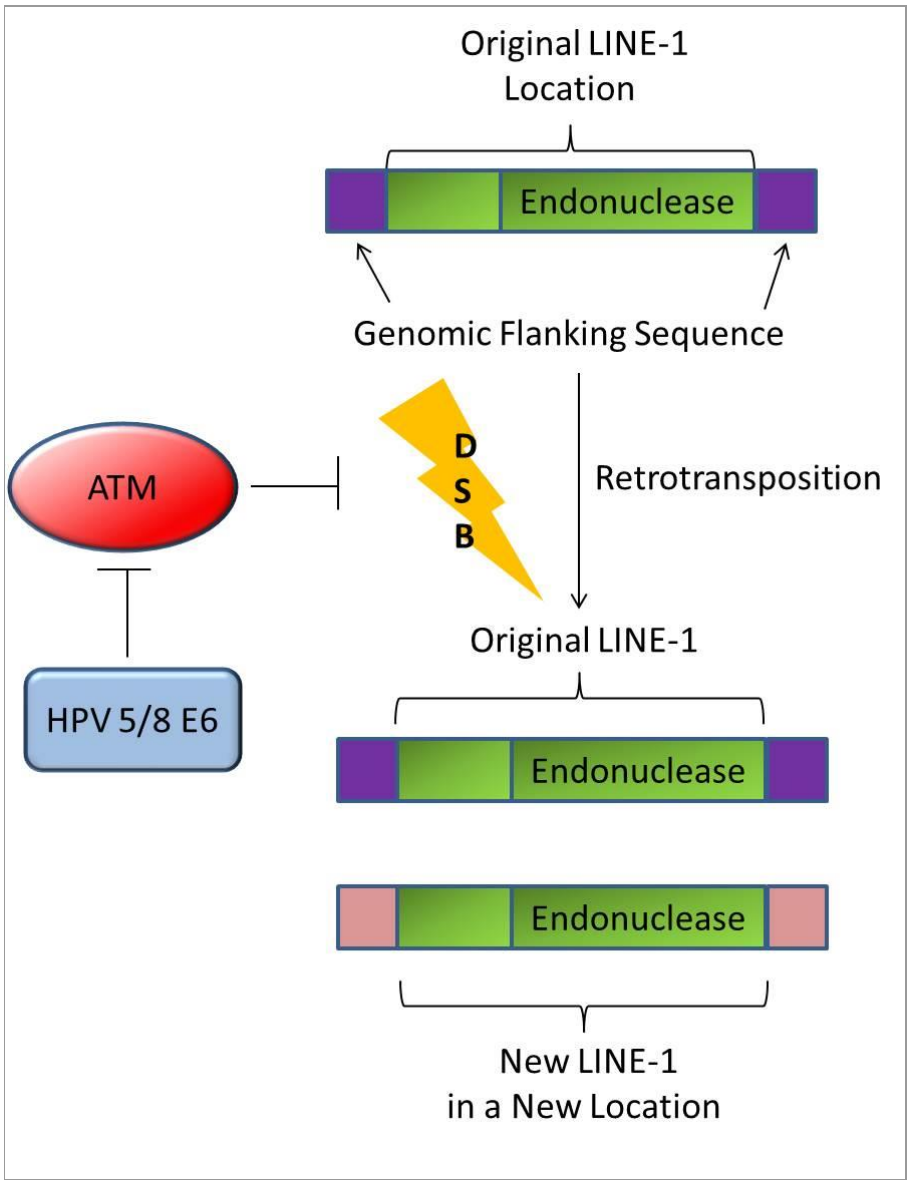

Image courtesy Dr. Nicholas Wallace.

β-HPV E6 protein attenuates LINE-1 retrotransposition. β-HPV E6 inhibits double-stranded break (DSB) repair by reducing ATM expression. ATM reduction also inhibits LINE-1 retrotransposition, but not LINE-1 induced DSBs, suggesting a potential mechanism for mutagenesis in β-HPV infected cells.