

Transcriptional Regulation Activities of EBV DNA Polymerase Processivity Factor BMRF1 on Viral Gene Expression

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Background/Objective

Epstein-Barr virus (EBV) is an important human pathogen that can persist in the host through latency and periodic reactivation. During lytic cycle, the viral DNA polymerase processivity factor BMRF1 associates with viral DNA polymerase BALF5 to promote lytic DNA replication. In addition to accessory activity, BMRF1 also functions as a transactivator to regulate promoter activities on cellular gastrin, viral OriLyt BHLF1 and BALF2 promoters. The aim was to approach the transcription regulatory function of BMRF1 in a whole virus setting.

Method

The BMRF1 deletion Maxi-EBV (p2089delBMRF1) was generated by PCR targeting. Maxi-EBV system and a DNA microarray containing 85 EBV genes were used to determine BMRF1-regulated gene candidates. In addition, possible BMRF1-interacting proteins which may participate in BMRF1-mediated transcription were analyzed by immunoprecipitation-mass spectrometry analysis.

Result

In EBV microarray analysis, BMRF1 regulated candidate genes included two early genes, BORF2 (ribonucleotide reductase 1) and BaRF1 (ribonucleotide reductase 2), and six late genes, BLLF1 (gp350/220), BLLF2 (function unknown), BILF2 (gp78/55), BcLF1 (major capsid protein), BDLF2 (cyclin B homolog) and BDLF3 (gp150). In luciferase reporter assays, we found BMRF1 turned on transcription activation on BcLF1, BLLF1, BLLF2 and BDLF3 promoter activities, but not on BILF2 promoter. Knockdown of SP1 decreased BMRF1-mediated activation on BDLF3 promoter, suggesting BMRF1 regulate BDLF3 expression through an SP1-dependent pathway. According to immunoprecipitation-mass spectrometry analysis, BMRF1 associates with chromatin modifiers, DNA replication/repair and RNA splicing association factors. Moreover, BMRF1 interacts with chromatin modifier BRG1 in the cells and in vitro. Knockdown of BRG1 reduced transactivation activity of BMRF1 on BHLF1, BcLF1 and BLLF1 promoters, suggesting BRG1 participates in the regulation of BMRF1 on a subset of viral gene expressions.

Conclusion

BMRF1 activates a subset of late promoters via cooperation with cellular transcription factor SP1 or chromatin modifier BRG1.