Deletion of Viral microRNAs in the Oncogenesis of Epstein–Barr Virus-Associated Lymphoma

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Epstein–Barr virus (EBV), which encodes >80 genes and nearly 50 non-coding RNAs, is a double-stranded DNA virus. EBV is associated with various types of lymphomas and lymphoproliferative disorders not only of B-cell but also T/NK-cell origin. However, the oncogenic mechanism remains poorly understood, including the EBV receptors expressed on T/NK cells, relationship of EBV with host genes, and epigenetic regulation of EBV and host genes. The roles of host and viral non-coding RNAs during tumorigenesis have been elucidated. EBV encodes at least 49 mature microRNAs (miRNAs), of which 44 are located in BamHI-A rightward transcripts (BARTs) region, and the remaining five are located in BamHI-H rightward fragment 1. BART miRNAs modulate cell differentiation, proliferation, apoptosis, and the cell cycle, and they are considered positive regulators of oncogenesis. We and others have recently reported that EBV-positive lymphomas frequently possess large deletions in BART miRNA clusters, suggesting that some viral miRNAs have suppressive effects on oncogenesis, and that deletion of these miRNAs may aid lymphoma formation.

Keywords: EBV, microRNA, BART, lymphomagenesis, CAEBV, ENKTL, diffuse large B cell lymphoma

INTRODUCTION

Epstein–Barr virus (EBV), which is the first human oncovirus, was isolated from Burkitt lymphoma by Epstein et al. (1964). EBV is a 170–180 kb double-stranded DNA virus belonging to the herpesvirus family and gammaherpesvirus subfamily, and its genome encodes approximately 80 genes (Longnecker et al., 2013). EBV belongs to the same subfamily as Kaposi’s sarcoma-associated herpesvirus, which is the causative virus of Kaposi’s sarcoma, and both viruses infect B cells and are related to B-cell lymphomas. In addition to Burkitt lymphoma, EBV is associated with a variety of malignancies with B-cell origins, such as Hodgkin lymphoma, diffuse large B-cell lymphoma (DLBCL), and immunodeficiency-related lymphoproliferative disorders detected after organ/hematopoietic stem cell transplantation (Swerdloff et al., 2016).

Epstein–Barr virus is also associated with natural killer (NK)-cell and T-cell neoplastic diseases including extranodal NK/T-cell lymphoma, nasal type (ENKTL) (Chan et al., 2017), chronic active EBV disease (CAEBV) (Quintanilla-Martinez et al., 2017; Cohen et al., 2020), and epithelial tumors such as nasopharyngeal carcinoma and gastric cancer (Cohen, 2000; Longnecker et al., 2013). Although extensive studies have been conducted on the implication of MYC translocation and

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activation in Burkitt lymphoma (Longnecker et al., 2013),
the oncogenic mechanisms (including the T/NK-cell receptors
expressed, the relationship of EBV with host genes, and epigenetic
regulation of EBV and host genes) associated with other
lymphoid tumors are still unclear.

The roles of host and viral non-coding RNAs have
been elucidated during tumorigenesis in various malignancies,
including EBV-associated diseases (Cai et al., 2006; Qiu et al.,
2011; Klinke et al., 2014; Kuzembayeva et al., 2014; Dreyfus, 2017;
Iizasa et al., 2020). We and others have recently reported that
EBV-positive lymphomas frequently possess large deletions in
viral microRNA (miRNA) clusters, suggesting that some miRNAs
negatively regulate lymphomagenesis (Peng et al., 2018; Okuno
et al., 2019; Mabuchi et al., 2021). This review outlines the role
of viral non-coding RNAs in the lymphomagenesis of EBV-
associated diseases, focusing on EBV-encoded miRNAs.

Biology of EBV Infection
Epstein–Barr virus can cause either a latent or lytic infection.
In a latent infection, EBV exists as an episome in the nucleus.
No viral particles are produced in the host cell, and only a
limited number of genes are expressed. Latent infections are
classified into four patterns, type 0, I, II, and III, depending
on the host cell, tissue, and immune status (Longnecker et al.,
2013; Munz, 2019). Latency type 0 is seen in the memory B cells
of healthy individuals, and few viral proteins are expressed in
this type. In type I, EBV nuclear antigen (EBNA) 1 and EBV-
encoded small RNAs (EBERs) are expressed. In a type II infection,
latent membrane protein (LMP) 1 and LMP2 are additionally
expressed. EBNA2, 3s, and LP are expressed in type III infections.

In contrast, immediate-early, early, and late genes are
expressed one after another during lytic infection, and virus
particles are produced. It was previously considered that virus-
infected cells are immediately in a latent state after EBV infects
B cells. However, recent evidence suggests that these cells are
temporarily in an abortive lytic infection state, and that the
expression of EBV lytic genes plays an important role in the
immortalization of infected cells (Ma et al., 2011; Murata et al.,
2014; Munz, 2019). In fact, these lytic genes are expressed in
hydroa vacciniforme-like lymphoproliferative disorder and
dLBCI (Yamamoto et al., 2016; Cohen et al., 2018), which
suggests that the abortive lytic infection may be involved in
lymphomagenesis.

Oncogenesis of EBV and EBV-Related
Malignancies
Epstein–Barr virus is used to immortalize human B cells
in vitro to produce a lymphoblastoid cell line (LCL). LMP1 is
a viral membrane oncprotein essential for immortalization;
it mimics CD40 expressed by T cells and constantly activates
downstream NF-κB, PI3K/AKT, JNK, and p38/MAPK pathways,
immortalizing infected cells and suppressing apoptosis (Kanda,
2018). Although LMP2A is not essential for immortalization, it
is also an oncprotein that mimics the B-cell receptor expressed
by B cells, resulting in constitutive calcium recruitment,
protein kinase C activation, cell proliferation, and differentiation
suppression (Longnecker et al., 2013). EBNA1, EBNA2, EBNA3A,
EBNA3C, and EBNA-LP are nuclear proteins that help
transform B cells and maintain latency (Tomkinson et al.,
1993; Kanda, 2018). All these latent infection-related genes
are expressed in a type III infection where host cell-mediated
immunity is suppressed, such as immunodeficiency-related
lymphoproliferative disorders. As only a limited number of
viral genes are expressed during latent type I and II infections,
it is easier to avoid host immunity with these infections
compared with a type III infection. However, immortalization
and suppression of apoptosis by viral oncoproteins are limited in
these latency types (Kimura, 2018).

EBV Non-coding RNAs
In addition to viral proteins, EBV encodes many non-coding
RNAs that also potentiate oncogenesis. EBER1 and EBER2,
which are long non-coding RNAs of 167 and 172 nucleotides,
respectively, are expressed most abundantly in EBV latency
infected cells at 10^7 copies per cell (Arrand and Rymo, 1982;
Howe and Shu, 1989). EBERs interact with a variety of RNA-
binding proteins and optimize B-cell transformation (Yajima
et al., 2005; Fok et al., 2006). However, the exact role of EBERs
is still unknown (Munz, 2019). EBV also encodes at least 49 mature
miRNAs. Of these 44 are located within the intronic regions
of type I and type II latency, in which a limited number of viral genes are expressed and evasion of
host immunity is easier (Longnecker et al., 2013). Notably, some
EBV-positive tumors have uneven distributions in specific areas
as shown in Table 1; whether this is due to genetic variations in
the host or differences in the infecting strains remains unclear.
miR-BART1-3p and miR-BART16 target CASP3 and inhibit apoptosis (Vereide et al., 2014), BIM, a pro-apoptotic protein of the bcl2 family, is targeted by several BART miRNAs (Marquitz et al., 2011). Another BART miRNA (miR-BART5-5p) targets PUMA to promote survival of host cells (Choy et al., 2008). BART miRNAs also help establish EBV infection and transformation by modulating viral and host functions in B cells. Both miR-BART1-3p and miR-BART1-5p suppress adaptive immunity mediated by CD4+ T cells by targeting IL12B and LY75 (Skalsky et al., 2012; Vereide et al., 2014; Tagawa et al., 2016). CD8+ T cell responses are also modulated by miR-BART1-3p and miR-BART17-5p which target IFI30 and TAP2, respectively (Albanese et al., 2016). Thus, BART miRNAs enhance lymphomagenesis. Similarly, BART miRNAs potentiate tumorigenesis in epithelial cells (Cai et al., 2015; Kanda et al., 2015; Qiu et al., 2015; Yang et al., 2017). The notion that BART miRNAs positively regulate oncogenesis is generally accepted.

In DNA viruses such as herpesviruses, exosomes released from virus-infected cells contain virus-derived components that contribute to the growth of the virus itself and the establishment of viral infections by regulating the host's immune system (Ansari et al., 2013). LMP1, which is an oncprotein bound to cytoplasmic membranes, is incorporated into exosomes, and plays a role in cancer progression (Nanbo et al., 2013; Sato et al., 2017). EBV miRNAs are also released from infected cells via exosomes to regulate uninfected adjacent cells and promote their growth, which contributes to tumorigenesis (Pegtel et al., 2010; Higuchi et al., 2018; Nanbo et al., 2018; Nkosi et al., 2020).

### Deletion of BART miRNA in EBV-Related Lymphomas

It has been reported that the lack of certain genes in human T-cell leukemia virus type 1, human papillomavirus, and Merkel cell polyomavirus results in increased tumorigenicity (Narisawa-Saito and Kiyono, 2007; Moore and Chang, 2010; Kataoka et al., 2015). In contrast, only a few studies have reported specific gene deletions in EBV-associated malignancies (Alfieri et al., 2015). In this next-generation sequencing era, whole viral genomes are easily sequenced directly from patient samples. We performed whole EBV sequencing by the hybrid capture method using 17,237 probes covering the entire EBV genome in EBV-infected peripheral blood and tumor tissues from patients with various EBV-associated diseases (Okuno et al., 2019; Mabuchi et al., 2014). Interestingly, 22 of 77 cases (35%) of CAEBV, which is a T- or NK-cell lymphoproliferative disease (Kimura et al., 2012; Kimura and Cohen, 2017; Quintanilla-Martinez et al., 2017), had a deletion of 73–49,847 bases in the EBV genome. A similar deletion was found in ENKTL (43%) and EBV-positive DLBCL (71%), which are lymphomas of NK- and B-cell origins, respectively. However, this intragenic deletion was not observed in infectious mononucleosis or post-transplant lymphoma.
suggesting that this deletion is a common phenomenon in certain types of EBV-positive lymphoma. The rarity of the intragenic deletion has been reported in healthy individuals and infectious mononucleosis patients (Palser et al., 2015; Yajima et al., 2021). In addition, EBV deletions were concentrated in the BART miRNA cluster regions (Figure 1). The most frequently deleted miRNAs were mir-BART6-5p and mir-BART6-3p, both of which negatively regulate the EBV immediate-early genes BZLF1 and BRLF1 (Iizasa et al., 2010). mir-BART18-5p and mir-BART20-5p also downregulate these immediate-early genes (Jung et al., 2014; Qiu and Thorley-Lawson, 2014). Mutated EBV lacking this region enhances lymphoma formation by inducing BZLF1 expression in xenograft models (Lin et al., 2015). Elevated BZLF1 due to loss of BART miRNAs may cause abortive lytic infection and promote lymphomagenesis (Munz, 2019).

In our genetic analysis, a group of lytic infection-related genes was also frequently missing outside the BART miRNA clusters, including core replication genes, which are essential for viral replication. We generated a mutant EBV strain lacking BALF5 (viral DNA polymerase catalytic subunit) (Narita et al., 2015) and used it to establish an LCL, which was then transplanted into immunodeficient mice (Okuno et al., 2019). The BALF5-deficient EBV produced lymphoma more frequently compared with the wild-type strain. Additionally, the BALF5-deficient LCL enhanced immediate-early/early gene expression, compared with the wild-type strain. These results suggest that deletion of BALF5, which is a core replication gene, induces the expression of lytic infection-related genes triggered by the immediate-early gene BZLF1 and promotes lymphoma formation. Multiple lytic infection genes, such as BNRF1, BGLF5, and BALF3, are involved in host genome instability (Manners et al., 2018; Xiong et al., 2020), and BHRF1 (viral BCL-2 homolog) and BCRF1 (viral interleukin-10 homolog) also promote cell proliferation (Xu et al., 2001; Zuo et al., 2011). EBV lacking core replication genes cannot produce virus particles or complete a lytic infection, but they can induce an abortive lytic infection and promote oncolytic infection by expressing a viral lytic infection gene (Munz, 2019; Murata et al., 2020). Thus, defective EBV strains may have some advantages during lymphomagenesis.

Interestingly, B95-8, which is the most potent laboratory strain, lacks most of the BART region (Figure 1; Baer et al., 1984; Klinke et al., 2014). Similar large deletions, including BART miRNA clusters, have been reported in patients with ENKTL and Hodgkin lymphoma (Peng et al., 2018; Kawatsuki et al., 2020). However, these defective viruses are relatively rarely associated with epithelial tumors (Cancer Genome Atlas Research Network (CGARN), 2014; Lin et al., 2014). The roles of BART miRNAs may differ between lymphoid and epithelial malignancies. The expression patterns of BART miRNAs depend on the infected cell lineage, and their expression levels vary widely among tumor types, with a 13-fold-increase in nasopharyngeal carcinoma and eightfold increase in gastric cancer relative to LCL and Burkitt lymphoma (Qiu et al., 2011).

Although intragenic deletions involving BART miRNAs have been detected in one-third of CAEBV patients, high expression levels of BART miRNAs are also seen in other patients with...
Epstein–Barr virus is a large DNA virus that encodes >80 genes and nearly 50 non-coding RNAs. Each non-coding RNA performs multiple and different functions, and their expression is regulated differentially according to cell type and micro-environment. EBV miRNAs have various functions and play pivotal roles in oncogenesis. It is clear that some viral miRNAs suppress lymphomagenesis, and that deletion of these miRNAs promotes lymphoma formation. Further research is necessary to elucidate the full roles of EBV miRNAs in tumorigenesis.

**REFERENCES**

Albanese, M., Tagawa, T., Bouvet, M., Maliqi, L., Lutter, D., Hoser, J., et al. (2016). Epstein-Barr virus microRNAs reduce immune surveillance by virus-specific CD8+ T cells. *Proc. Natl. Acad. Sci. U.S.A.* 113, E6467–E6475. doi: 10.1073/pnas.1605884113

Alfieri, C., and Joncas, J. H. (1987). Biomolecular analysis of a defective nontransforming Epstein-Barr virus (EBV) from a patient with chronic active EBV infection. *J. Virol.* 61, 3306–3309.

Amoroso, R., Fitzsimmons, L., Thomas, W. A., Kelly, G. L., Rowe, M., and Bell, A. I. (2011). Quantitative studies of Epstein-Barr virus-encoded microRNAs provide novel insights into their regulation. *J. Virol.* 85, 996–1010. doi: 10.1128/JVI.01528-10

Ansari, M. A., Singh, V. V., Dutta, S., Veettil, M. V., Dutta, D., Chikoti, L., et al. (2013). Constitutive interferon-inducible protein 16-inflammasome activation during Epstein-Barr virus latency I, II, and III in B and epithelial cells. *J. Virol.* 87, 8606–8623. doi: 10.1128/JVI.00805-13

Arrand, J. R., and Rymo, L. (1982). Characterization of the major Epstein-Barr virus-specific RNA in Burkitt lymphoma-derived cells. *J. Virol.* 41, 376–389. doi: 10.1128/JVI.41.2.376-389.1982

Baer, R., Bankier, A. T., Biggin, M. D., Deininger, P. L., Farrell, P. J., Gibson, T. J., et al. (1984). DNA sequence and expression of the B95-8 Epstein-Barr virus genome. *Nature* 310, 207–211.

Bernhardt, K., Haar, J., Tsai, M. H., Poirey, R., Feederle, R., and Delecluse, H. J. (2016). A viral microRNA cluster regulates the expression of PTEN, p27 and of a bcl-2 homolog. *PLoS Pathog.* 12:e1005405. doi: 10.1371/journal.ppat.1005405

Cai, L. M., Lyu, X. M., Luo, W. R., Cui, X. F., Ye, Y. F., Yuan, C. C., et al. (2015). EBV-miR-BART7-3p promotes the EMT and metastasis of nasopharyngeal carcinoma cells by suppressing the tumor suppressor PTEN. *Oncogene* 34, 2156–2166. doi: 10.1038/onc.2014.341

Cai, X., Schafer, A., Lu, S., Bijelei, J. P., Desrosiers, R. C., Edwards, R., et al. (2006). Epstein-Barr virus microRNAs are evolutionarily conserved and differentially expressed. *PLoS Pathog.* 2:e23. doi: 10.1371/journal.ppat.0020023

Cancer Genome Atlas Research Network (CGARN) (2014). Comprehensive molecular characterization of gastric adenocarcinoma. *Nature* 513, 202–209. doi: 10.1038/nature13480

Chan, J. K. C., Quintanilla-Martinez, L., and Ferry, J. A. (2017). “Extranodal NK/T-cell lymphoma, nasal type,” in *WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues*, revised 4th Edn, eds S. H. Swerdlow, E. Campo, N. L. Harris, E. S. Jaffe, S. A. Pileri, H. Stein, et al. (Lyon: WHO IARC Press), 368–371.

Chen, Y., Fachko, D., Ivanov, N. S., Skinner, C. M., and Skalsky, R. L. (2019). Epstein-Barr virus microRNAs regulate B cell receptor signal transduction and lytic reactivation. *PLoS Pathog.* 15:e1007535. doi: 10.1371/journal.ppat.1007535

Choy, E. Y., Siu, K. L., Kok, K. H., Lung, R. W., Tsang, C. M., To, K. F., et al. (2008). An Epstein-Barr virus-encoded microRNA targets PUMA to promote host cell survival. *J. Exp. Med.* 205, 2551–2560. doi: 10.1084/jem.20072581

Cohen, J. I. (2000). Epstein-Barr virus infection. *N. Engl. J. Med.* 343, 481–492.

Cohen, J. I., Iwatsuki, K., Ko, Y. H., Kimura, H., Manoli, I., Ohshima, K., et al. (2020). Epstein-Barr virus NK and T cell lymphoproliferative disease: report of a 2018 international meeting. *Leuk Lymphoma* 61, 808–819. doi: 10.1080/10428194.2019.1699080

Cohen, M., Vistarop, A. G., Huanan, F., Narbaizt, M., Metrebian, F., De Matteo, E., et al. (2018). Epstein-Barr virus lytic cycle involvement in diffuse large B cell lymphoma. *Hematol. Oncol.* 36, 98–103. doi: 10.1002/hon.2465

Dreyfus, D. H. (2017). Genetics and molecular biology of Epstein-Barr Virus-Encoded BART MicroRNA: a paradigm for viral modulation of host immune response genes and genome stability. *J. Immunol. Res.* 2017:4758539. doi: 10.1155/2017/4758539

Epstein, M. A., Achong, B. G., and Barr, Y. M. (1964). Virus particles in cultured virus-infected lymphoblasts from Burkitt's lymphoma. *Lancet* 373, 702–703. doi: 10.1016/s0140-6736(64)91524-7

Fink, S. E., Gandhi, M. K., Nourse, J. P., Keane, C., Jones, K., Crooks, P., et al. (2014). A comprehensive analysis of the cellular and EBV-specific microRNAome in primary CNS PTLD identifies different patterns among EBV-associated tumors. *Am. J. Transplant.* 14, 2577–2587. doi: 10.1111/ajt.12858

Fok, V., Friend, K., and Steitz, J. A. (2006). Epstein-Barr virus noncoding RNAs undergo nucleocytoplasmic shuttling. *J. Cell. Biol.* 173, 319–325. doi: 10.1083/jcb.200601026

Haneklaus, M., Gerlic, M., Kurowska-Stolarska, M., Rainey, A. A., Pich, D., McNees, I. B., et al. (2012). Cutting edge: miR-223 and EBV miR-BART15 performs multiple and different functions, and their expression is regulated differentially according to cell type and micro-environment. EBV miRNAs have various functions and play pivotal roles in oncogenesis. It is clear that some viral miRNAs suppress lymphomagenesis, and that deletion of these miRNAs promotes lymphoma formation. Further research is necessary to elucidate the full roles of EBV miRNAs in tumorigenesis.

**AUTHOR CONTRIBUTIONS**

All authors contributed to the concept development process and to the writing and review of the manuscript and also gave final approval of the version to be published.

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regulate the NLRP3 inflamasome and IL-1beta production. J. Immunol. 189, 3795–3799. doi: 10.4049/jimmunol.1200312
Higuchi, H., Yamakawa, N., Imadome, K. I., Yahata, T., Kotaki, R., Ogata, J., et al. (2018). Role of exosomes as a proinflammatory mediator in the development of EBV-associated lymphoma. Blood 131, 2552–2567. doi: 10.1182/blood-2017-07-794529
Hooykaas, M. J. G., van Gent, M., Poppe, J. A., Kruse, E., Boer, I. G. J., van Leenhen, D., et al. (2017). EBV microrna BART16 suppresses Type I IFN signaling. J. Immunol. 198, 4062–4075. doi: 10.4049/jimmunol.1501605
Howe, J. G., and Shu, M. D. (1989). Epstein-Barr virus small RNA (EBER) genes: unique transcription units that combine RNA polymerase II and III promoter elements. Cell 57, 825–834. doi: 10.1016/0092-8674(89)90797-6
Huang, W. T., and Lin, C. W. (2014). EBV-encoded mi-BART20-5p and miR-BART8 inhibit the IFX-gamma-ST1 pathway associated with disease progression in nasopharyngeal carcinoma. Am. J. Pathol. 184, 1185–1197. doi: 10.1016/j.ajpath.2013.12.024
Iizasa, H., Kim, H., Kartika, A. V., Kanehiro, Y., and Yoshiyama, H. (2020). Role of viral and host microRNAs in immune regulation of Epstein-Barr virus-associated diseases. Front. Immunol. 11:367. doi: 10.3389/fimmu.2020.00336
Iizasa, H., Wulf, B. E., Alla, N. R., Maragakis, M., Megraw, M., Hatzigeorgiou, A., et al. (2010). Editing of Epstein-Barr virus-encoded BART miRNAs controls their dicer targeting and consequently affects viral latency. J. Biol. Chem. 285, 33358–33370. doi: 10.1074/jbc.M110.138362
Jung, Y. J., Choi, H., Kim, H., and Lee, S. K. (2014). MicroRNA miR-BART20-5p stabilizes Epstein-Barr virus latency by directly targeting BZLF1 and BRLF1. J. Virol. 88, 9027–9037. doi: 10.1128/JVI.01721-14
Kanda, T. (2018). EBV-encoded latent genes. Adv. Exp. Med. Biol. 70, 1032–1033. doi: 10.1007/978-3-319-43415-8_17
Kawano, Y., Iwata, S., Kawada, J., Gotoh, K., Suzuki, M., Torii, Y., et al. (2013). Plasma viral microRNA Profiles reveal potential biomarkers for chronic active Epstein-Barr virus infection. J. Infect. Dis. 208, 771–779. doi: 10.1093/infdis/jit222
Kawatsuki, A., Igawa, T., Urata, T., Tanaka, T., Sato, Y., and Yoshino, T. (2020). Deletion of BART miRNA-encoding cluster in Epstein-Barr virus DNA in classic Hodgkin lymphoma. Pathol. Int. 70, 1032–1033. doi: 10.1111/pin.13022
Kimura, H. (2018). EBV in T-NK-Cell Tumorigenesis. Adv. Exp. Med. Biol. 1045, 377–394. doi: 10.1007/978-981-10-7230-7_17
Kanduri, K., Miyata, M., Kano, M., Kondo, S., Yoshizaki, T., and Iizasa, H. (2015). Clustered microRNAs of the Epstein-Barr virus cooperatively downregulate an epithelial cell-specific metastasis suppressor. J. Virol. 89, 2684–2697. doi: 10.1128/JVI.03189-14
Katoaka, K., Nagata, Y., Kitakama, A., Shiraiishi, Y., Shimamura, T., Yasunaga, I., et al. (2015). Integrated molecular analysis of adult T-cell leukemia/lymphoma. Nat. Genet. 47, 1304–1315. doi: 10.1038/ng.3415
Kataoka, M., Nagata, Y., Kitanaka, A., Shiraishi, Y., Shimamura, T., Y asunaga, J., et al. (2020). Role of viral and host microRNAs in immune regulation of Epstein-Barr virus-infected cells. J. Virol. 87, 10334–10347. doi: 10.1128/JVI.01310-13
Narisawa-Saito, M., and Kiyono, T. (2007). Basic mechanisms of high-risk human papillomavirus-induced carcinogenesis: roles of E6 and E7 proteins. Cancer Sci. 98, 1505–1511. doi: 10.1111/j.1349-7006.2007.00546.x
Narita, Y., Sugimoto, A., Kawashima, D., Watanabe, T., Kanda, T., Kimura, H., et al. (2015). A Herpesvirus specific motif of Epstein-Barr Virus DNA polymerase is required for the efficient lytic genome synthesis. Sci. Rep. 5:11767. doi: 10.1038/srep11767
Nkosi, D., Sun, L., Duke, L. C., Patel, N., Surapaneni, S. K., Singh, M., et al. (2020). Epstein-Barr Virus LMP1 promotes Syntenin-1- and Hrs-Induced extracellular vesicle formation for its own secretion to increase cell proliferation and migration. mBio 11, e589–e520. doi: 10.1128/mBio.00589-20
Narita, Y., Sugimoto, A., Kawashima, D., Watanabe, T., Kanda, T., Kimura, H., et al. (2015). A Herpesvirus specific motif of Epstein-Barr Virus DNA polymerase is required for the efficient lytic genome synthesis. Sci. Rep. 5:11767. doi: 10.1038/srep11767
Pegtel, D. M., Cosmopoulos, K., Thorley-Lawson, D. A., van Eijndhoven, M. A., Hopmans, E. S., Lindenberg, J. L., et al. (2010). Functional delivery of viral miRNAs via exosomes. Proc. Natl. Acad. Sci. U.S.A. 107, 6328–6333. doi: 10.1073/pnas.091843107
Peng, R. J., Han, B. W., Cai, Q. Q., Zuo, X. Y., Xia, T., Chen, J. R., et al. (2018). Genomic and transcriptionic landscapes of Epstein-Barr virus in extranodal natural killer T-cell lymphoma. Leukemia 33, 1451–1462. doi: 10.1038/s41375-018-0324-5
Qiu, J., Cosmopoulos, K., Pegtel, M., Hopmans, E., Murray, P., Middeldorp, J., et al. (2011). A novel persistence associated EBV miRNA expression profile is disrupted in neoplasia. PLoS Pathog. 7:e1002193. doi: 10.1371/journal.ppat.1002193
Qiu, J., Smith, P., Leahy, L., and Thorley-Lawson, D. A. (2015). The Epstein-Barr virus encoded BART miRNAs potentiate tumor growth in vivo. PLoS Pathog. 11:e1004561. doi: 10.1371/journal.ppat.1004561

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Kimura, R. L., Corcoran, D. L., Gottwein, E., Frank, C. L., Kang, D., Hafner, M., Sakamoto, K., Sekizuka, T., Uehara, T., Hishima, T., Mine, S., Fukumoto, H., Qiu, J., and Thorley-Lawson, D. A. (2014). EBV microRNA BART 18-5p targets Kimura et al. EBV miRNA in Oncogenesis Swerdlow, S. H., Campo, E., Pileri, S. A., Harris, N. L., Stein, H., Siebert, R., Quintanilla-Martinez, L., Ko, Y. H., Kimura, H., and Jaffe, E. S. (2017). “EBV– Seto, E., Moosmann, A., Gromminger, S., Walz, N., Grundhoff, A., and Hammerschmidt, W. (2010). Micro RNAs of Epstein-Barr virus promote cell cycle progression and prevent apoptosis of primary human B cells. PLoS Pathog. 6:e1001063. doi: 10.1371/journal.ppat.1001063 Skalsky, R. L., Corcoran, D. L., Gottwein, E., Frank, C. L., Kang, D., Hafner, M., et al. (2012). The viral and cellular microRNA targetome in lymphoblastoid cell lines. PLoS Pathog. 8:e1002484. doi: 10.1371/journal.ppat.1002484 Swerdlow, S. H., Campo, E., Pileri, S. A., Harris, N. L., Stein, H., Siebert, R., et al. (2016). The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood 127, 2375–2390. doi: 10.1182/blood-2016-01-643569 Tagawa, T., Albanese, M., Bouvet, M., Moosmann, A., Mautner, J., Heissmeyer, V., et al. (2016). Epstein-Barr viral miRNAs inhibit antiviral CD4+ T cell responses targeting IL-12 and peptide processing. J. Exp. Med. 213, 2065–2080. doi: 10.1084/jem.20160248 Tomkinson, B., Robertson, E., and Kieff, E. (1993). Epstein-Barr virus nuclear proteins EBNA-3A and EBNA-3C are essential for B-lymphocyte growth transformation. J. Virol. 67, 2014–2025. doi: 10.1128/JVI.67.4.2014-2025.1993 van Eindhoven, M. A., Zajistra, J. M., Groenewegen, N. J., Drees, E. E., van Nice, S., Baglio, S. R., et al. (2016). Plasma vesicle miRNAs for therapy response monitoring in Hodgkin lymphoma patients. JCI Insight 1:e89631. doi: 10.1172/jci.insight.89631 Vereide, D. T., Seto, E., Chiu, Y. F., Hayes, M., Tagawa, T., Grundhoff, A., et al. (2014). Epstein-Barr virus maintains lymphomas via its miRNAs. Oncogene 33, 1258–1264. doi: 10.1038/onc.2013.71 Xia, T., O’Hara, A., Araujo, I., Barreto, J., Carvalho, E., Sapucaia, J. B., et al. (2008). EBV microRNAs in primary lymphomas and targeting of CXCL-11 by ebv-mir-BHRF1-3. Cancer Res. 68, 1436–1442. doi: 10.1158/0008-5472.CAN-07-5126 Xiong, J., Cui, B. W., Wang, N., Dai, Y. T., Zhang, H., Wang, C. F., et al. (2020). Genomic and transcriptomic characterization of natural killer T cell lymphoma. Cancer Cell 37, 403.e6–419.e6. doi: 10.1016/j.ccell.2020.02.005 Xu, Z. G., Iwatsuki, K., Oyama, N., Ohitsuka, M., Satoh, M., Kikuchi, S., et al. (2001). The latency pattern of Epstein-Barr virus infection and viral IL-10 expression in cutaneous natural killer/T-cell lymphomas. Br. J. Cancer 84, 920–925. Yajima, M., Kakuta, R., Saito, Y., Kitaya, S., Toyoda, A., Ikuta, K., et al. (2021). A global phylogenetic analysis of Japanese tonsil-derived Epstein-Barr virus strains using viral whole-genome cloning and long-read sequencing. J. Gen. Virol. 102:001549. doi: 10.1099/jgv.0.001549 Yajima, M., Kanda, T., and Takada, K. (2005). Critical role of Epstein-Barr Virus (EBV)-encoded RNA in efficient EBV-induced B-lymphocyte growth transformation. J. Virol. 79, 4298–4307. Yamamoto, T., Hirai, Y., Miyake, T., Hamada, T., Yamasaki, O., Morizane, S., et al. (2016). Epstein-Barr virus reactivation is induced, but abortive, in cutaneous lesions of systemic hydroa vacciniforme and hypersensitivity to mosquito bites. J. Dermatol. Sci. 82, 153–159. doi: 10.1016/j.jdermsci.2016.03.001 Yang, Y. C., Liem, A., Lambert, P. F., and Sugden, B. (2017). Dissecting the regulation of EBV’s BART miRNAs in carcinomas. Virology 505, 148–154. doi: 10.1016/j.virol.2017.02.013 Zuo, J., Thomas, W. A., Haigh, T. A., Fitzsimmons, L., Long, H. M., Hislop, A. D., et al. (2011). Epstein-Barr virus evades CD4+ T cell responses in lytic cycle through BLZF1-mediated downregulation of CD74 and the cooperation of vBcl-2. PLoS Pathog. 7:e1002455. doi: 10.1371/journal.ppat.1002455 Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.