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Epstein Barr Virus's Role in Cancer and Its Association with the p53 Family in Inhibiting Apoptosis

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Abstract:- Epstein Barr Virus (EBV), belonging to the family of herpes virus, widely responsible for initiating a long-lasting infection that infects more than 90% of the adult human population world-wide. Apart from being a chronic B-Lymphocyte infecting virus with common symptoms associated with fever, sore throat, lymphadenopathy etc., the viral proteins have been reported to be linked to different types of human carcinomas & lymphomas in the long run. The approach of this review is mostly to bring forward the insights of some of the potent carcinomas that are directly related to the pathogenesis of this virus, elucidating the viral proteins -: EBNA-1, EBNA-2, EBNA-LP, EBNA-3A, 3B, 3C, LMP-1, their direct association with human p53, p63, p73 & their isoforms in regulating the tumour inducing pathways, along with this some of the recent therapeutic approaches in preventing the EBV linked carcinogenesis has also been incorporated in this review.

Keywords:- *Epstein Barr Virus (EBV), p53, p63, p73, EBNA, LMP, Lymphoma, Malignancy, CRISPR, CAR-T Cell.*

I. INTRODUCTION

Epstein Barr Virus (EBV) is an omnipresent virus also known as Human gamma herpesvirus 4. The International Agency for Research on Cancer (IARC) classified it as a Category I carcinogen after discovering it in cultured tumour cells taken from Burkitt's lymphoma biopsy in 1964 [1], EBV has the same structure as other herpesviruses. The viral genome is present as a linear, double-stranded, ca.172-kbp DNA molecule in the infectious virion. The viral genome is mostly preserved as an extrachromosomal episome in infected cells [1]. Around 90% of the world population are EBV positive, Preadolescent infections are typically asymptomatic, but primary infections later in life commonly result in Infectious Mononucleosis (IM) [2]. Following a primary infection, the EBV enters a latency phase in which it attacks epithelial cells, enters the circulating B lymphocyte, and remains in a dormant condition for the rest of one's life, similar to other herpesviruses [3]. Latent EBV infection contributes to 2% of all human malignancies, including numerous Nasopharyngeal Carcinomas (NPCs), Burkitt's Lymphomas (BLs), and ~10% of gastric cancers [2] EBV was the first human cancer virus discovered, and it causes around 1.8 percent of all malignancies in humans, including Hodgkin lymphoma, Burkitt's lymphoma, NK/T cell lymphoma, and others [4]. EBV infection has been observed in tumour cells of Nasopharyngeal Carcinoma (NPC) patients from endemic regions, and it has recently been linked to Gastric Cancer (GC), where EBV infection has been identified in roughly 10% of all cases [5]. The oral route is the most common route for EBV to spread. Organ transplantation and blood transfusion, have been associated to the spread of EBV [3]. EBV initially infects the oropharyngeal epithelial cell, followed by the establishment of the latency in B cells, epithelial cells and natural killer/T cells for its lifelong persistence, [6]. The virus, in general, does not cause any harm. However, in compromised health conditions, increased viral titre may lead to subsequent complications including various types of cancers [7], EBV has developed to have an uncanny ability to govern cell death. TP53, the gene encoding for the tumour suppressor protein p53, which is responsible for cell cycle arrest and apoptosis, is the most frequently mutated gene in human carcinomas [5]. Viral and bacterial infections strongly modulate TP53 mutation frequency due to its capability to interfere with p53 activity [8]. It has been found that several EBV gene products produced during lytic and latent phase tends to interact with P53 isoforms such as P63 and P73 causing mutations [9]. Also, Epigenetic modulation induced by EBV proteins, could be responsible for p53 deregulation [10]. Although rare TP53 mutations are seen in EBV-associated epithelial malignancies [11]. In Gastric Cancer the TP53 pathway deregulation is due to TP53 mutations in approximately 70% of all cases, but in the EBV-associated gastric cancers mutations are infrequent [5]. The review has focussed on the interactions of EBV with P53 family, and its potent role in human malignancies

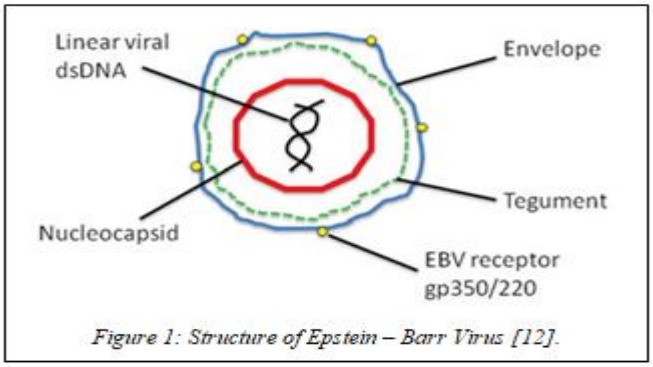

Figure 1: Structure of Epstein – Barr Virus [12].

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II. PATHOGENESIS OF EBV

Epstein-Barr Virus (EBV) led infectious mononucleosis is very much frequent among the adolescent and young adult groups of the western society. Due to its route of transmission, which is mostly through saliva, EBV infections are commonly referred to as "Kissing Disease." The EBV infection normally begins sub-clinically in children, developing a chronic infection in the B lymphocytes, thus producing a low-level viral load in the saliva, which can be easily spread through intimate contacts, sharing foods and utensils, cough, sneeze, etc. EBV has been reportedly found in both male and female genital fluids, thus indicating that the virus can also be transmitted through sexual contact [13]. The most common symptoms that are manifested by EBV positive population, include persistent fever, sore throat, fatigue, lymphadenopathy [14]. As a person gets infected with the EBV virus, the virus remains in a dormant condition within the resting memory B lymphocytes of the host for rest of the life. However, reportedly being a strong transforming virus for B cells in vitro, it is linked to a number of significant lymphomas, which include Burkitt's, Hodgkin's, and Immunoblastic lymphoma [15]. Two EBV subtypes have been found till date EBV-1 & EBV-2, these subtypes basically show a variation in their genetic makeup, other biological properties based on their geographical locations [16][17]. Considering the latent EBV genes, EBER1, EBER2, EBNA2, EBNA3, and EBNA-LP have the most genetic differences. EBV-1 predominates in most regions of the world around 80–90% infectivity rate, with EBV-2 found more frequently in equatorial Africa, New Guinea. This subtype is also very common among the immunocompromised or AIDS population [16]. After the initial exposure to the infection in the oral & nasal epithelial lining, the virus is expected to undergo a short phase of lytic replications [18][19][20]. Following the infection of naive B cells in adjacent tonsillar lymphoid tissues, a short “pre-latent” period of lytic and latent gene expression occurs before the epigenetic regulation of viral genes [21]. Limited expression of a restricted collection of lytic genes with regulatory function, thus omitting lytic genes needed for DNA replication and virion assembly, characterizes this short pre-latent phase. Pre-latent lytic gene products, such as BART mi-RNA, viral BCL-2 homologues, and BZLF1, are thought to contribute to the early survival of EBV – infected B cells by boosting cell proliferation and blocking apoptosis [22]. Rapid development of latently infected B cells is produced by expression of the complete growth-promoting complement of latency genes, i.e., the latency III program, after epigenetic suppression of the complete complement of lytic genes and a subset of latent gene promoters [23]. Infected epithelial cells and B cells express the entire complement of lytic and latent antigens, triggering a robust humoral and cellular immune response that suppresses viral replication [24][25][26]. B cells which had been latently infected, convert from the highly immunogenic latency III program to the less immunogenic latency II program, with viral gene expression limited to three proteins: EBNA-1, LMP-1, and LMP-2A [31]. The viral genome is maintained by EBNA-1 [27], while LMP-1 and LMP-2A, on the other hand, promote cell proliferation avoiding apoptosis [28][29]. Latency II B cells can adopt a germinal centre B-cell phenotype without EBNA-2-mediated trans-activation, allowing them to survive germinal centre and/or extra-follicular growth and maturation into EBV – infected memory B cells [30][31]. Memory B cells infected with EBV endure by transitioning from the latency II to the latency 0 program, with almost no viral gene expression and only sporadic LMP-2a expression [30][32][33]. Resting memory B cells infected with EBV circulate in the bloodstream, seeding lymphoid tissues all throughout the body. End-stage viral replication is caused by plasmacytic differentiation of EBV – positive memory B cells [30][34][35]. Intermittent virus replication in oral and nasal tissues also results in a low-level viral shedding in saliva, as well as lifelong survival of IgG anti-VCA antibody and EBV – specific cytotoxic T lymphocytes (CTL), which are more often directed against lytic antigens than latent antigens [36].

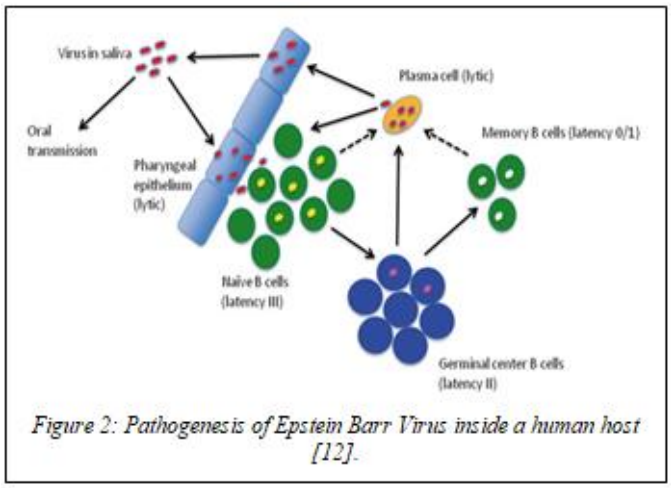

Figure 2: Pathogenesis of Epstein Barr Virus inside a human host [12].

III. EBV GENES RESPONSIBLE FOR CARCINOGENESIS

EBV encodes for fascinating genes. Anti-apoptotic chemicals, cytokines, and signal transducers interact with or have homology with these products, encouraging EBV infection, immortalization, and transformation. The genes contain various nuclear proteins (the Epstein Barr nuclear antigens, [EBNAs]), some plasma membrane proteins (the latent membrane proteins, [LMPs]) and also some very abundant small untranslated ribonucleic acids ([RNAs]. Depending on the cell type in which it dwells, the EBV virus might employ different transcription programmes. Latency I, III, and the lytic cycle are the three major gene expression programmes. Latency II shows intermediate gene expression [37].

- **EBNA – 1:**

EBNA-1 is a sequence-specific DNA binding phosphoprotein essential for EBV genome replication and maintenance. EBNA-1 binding to oriP serves as a transcription enhancer for the Cp EBNA promoter and the promoter for LMP-1 in addition to DNA replication [38, p. 1]. EBNA-1 interacts with the plasmid replication origin, which is made up of two different EBNA-1 binding regions [39] [40] [41]. They are the family of repeats and the dyad

symmetry [42]. Multiple 18-bp EBNA-1 binding sites are found in both the repeat family and the dyad symmetry binding elements [42]. EBV employs host enzymes to facilitate the remaining stages of replication after EBNA-1 binds to the plasmid origin of replication. As the family of repeats element aborts leftward replication, EBV genome replication only propagates rightward, forcing episomal replication to begin and end at the plasmid origin of replication.

- **EBNA – 2:**

EBNA-2 and EBNA-LP are the first latent proteins that are detected after EBV infection. EBNA-2 is a transcriptional co-activator that coordinates viral gene expression in latency III and also trans-activates various genes, and also plays a vital role for cell immortalization [43] [39]. Through interactions with the overexpression of cellular proteins, EBNA2 prevents intrinsic cell death. EBNA-2 can bind and inhibit the orphan nuclear receptor Nur77 [44][44], which has been reported to bind and affect the activity of many pro-survival BCL-2 family members. EBNA2 is involved in the upregulation of viral and cellular gene expression. CD23 (a surface marker of activated B cells), c-myc (a cellular proto-oncogene), and the viral EBNA-C promoter are among them [45]. The activation of myc, can both cause increase proliferation and sensitise cells to apoptosis, through long-range interactions [46].

- **EBNA – 5 / EBNA – LP:**

EBNA-LP, also known as EBNA-5, is one of the first viral proteins produced during EBV infection of B cells [43]. EBNA-LP is the transcriptional co-activator of EBNA2, is essential for efficient B cell transformation [46]. EBNA-LP tends to interacts with EBNA-2 to drive resting B lymphocytes into the G1 phase of the cell cycle by binding and inactivating cellular p53 and retinoblastoma protein tumour suppressor gene products [47, p. 2] [47, p. 2]. EBNA-LP is also found to interacts with other transcription factors involved in the Notch Signalling Pathway [47, p. 2] [47, p. 2].

- **EBNA-3A, EBNA-3B & EBNA-3C:**

EBNA-3A, EBNA-3B, and EBNA-3C act as transcriptional regulators, with repressing and activating properties [48]. Three adjacent genes on the viral genome encode members of the EBNA-3 family [43]. Both EBNA-3A and EBNA-3C have been found to be crucial for B-Cell immortalization [49][49]. EBNA-3C may bypass the retinoblastoma tumour suppressor gene checkpoint in the G1 phase of the cell cycle [50, p. 7]. EBNA3C is involved in upregulation of LMP-1[51]. EBNA3B has been found to induce expression of vimentin and CD40 [52, p. 4]. Cp binding factor interacts with all three EBNA-3s. 1. Cp binding factor 1 is a component of the notch signalling system, and overexpression of the notch protein has been linked to human T-cell malignancies [53].

- **LMP – 1:**

LMP-1 is a membrane protein having six membrane-spanning hydrophobic regions and a C-terminal cytoplasmic domain that appears to house the molecule's effector component. Through its C-terminal intra-cytoplasmic domains, LMP1 causes a persistent activation of numerous signal transduction cascades in human cells. As a result, LMP1 operates as a continually active receptor that is not triggered by external ligands [54]. LMP1 clustering is induced by transmembrane domains, which trigger signalling from a 200-residue cytoplasmic tail. Two LMP1 signalling domains were found to be important for EBV-mediated B-cell growth transformation in a reverse genetic investigation. These transformation effector site (TES)/C-terminal activation region (CTAR) 1 and 2, mimics CD40, a tumour necrosis factor (TNF) receptor family member and important B-cell co-stimulatory receptor. LMP1 activates the NF-B, mitogen-activated protein kinase (MAPK), phosphatidyl-inositol 3-kinase, and interferon regulatory factor (IRF) pathways in a constitutive manner. To limit migration and induce latency, the LMP1 CTAR3 domain triggers SUMOylation pathways [55].

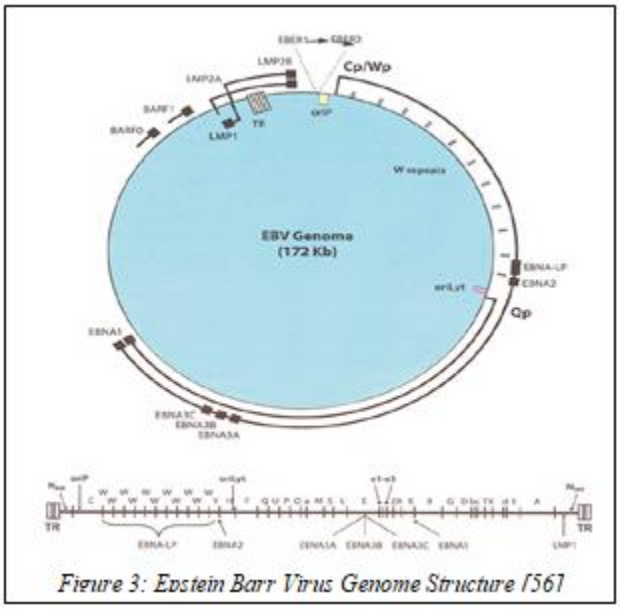

Figure 3: Epstein Barr Virus Genome Structure [56]

P53, P63, P73 THEIR ISOFORMS & THE INTERACTION WITH EBV

p53 gene is a type of tumour suppressor gene which is responsible for prevention of tumour formation in the body. This p53 gene has been located on chromosome 17 of the human genome. The encoded p53 protein binds to DNA in the cell, which causes another gene to generate the protein p21, which interacts with a cell division-stimulating protein (cdk2). When p21 binds to cdk2, the cell is unable to progress to the next stage of cell division resulting in apoptosis of the cell [57][58]. A mutation in the p53 gene prevents it from binding to the DNA in an effective manner, hence further the p21 protein could no longer act as a 'stop signal' for cell division. Thus, resulting in uncontrolled cell divisions and tumour growth [59]. Recently discovered, two other isoforms which includes the p63 & p73 protein which whole together make up the p53 family. These two proteins share a great extent of homologies, which came into existence as a result of the triplication of a commonly inherited gene [60]. The three shared structural domains of

p53, p63, and p73 that have been identified so far are the N-Terminal Transactivation Domain (TAD), DNA-Binding Domain (DBD), and the C-Terminal Oligomerization Domain (OD) [61][58]. P63 is involved in the development of the limbs, skin, and craniofacial region, while p73 is believed to aid neurogenesis [62][63]. The malfunctioning of any of the p53 family proteins has been linked to the advancement of cancer in several studies. Both structurally and functionally, the p63 and p73 proteins are identical to the p53 protein. P53 family members are expressed in a variety of isomeric forms due to alternate carboxyl-terminal splicing and the use of diverse promoters. P1 promoter-based transcription resulted in TA isoforms, which are the TAp63 & TAp73. While, P2 promoter-based transcription lacks the TA domain and the N-Terminal region remains truncated giving rise to the ΔN isoforms which are the ΔNp63 & ΔNp73 [64][65][66]. The activities of ΔN and TA isoforms as an anti-apoptotic factor and a pro-apoptotic factor, respectively, are further elaborated by the fact that N isoform acts as a crucial negative inhibitor of TA isoform [67].

- **EBV & p53 ISOFORM INTERACTIONS:**

In Non-Hodgkin's Lymphoma (NHL) of the head and neck, Nasopharyngeal cancer, Burkitt's lymphoma, and gastric carcinoma, a link between the EBV infection and p53 expression is observed [68]. When the human peripheral blood mononuclear cells were infected with the EBNA3C construct, it resulted in the deletion of residues 130–159 of the EBNA3C open reading frame (ORF) which changed p53 expression compared to wild type EBV [69]. The N-terminal domain of EBNA3C, with 130–190 residues has reportedly inhibited p53 transcriptional activity via decreasing p53 DNA binding activity, on performing a luciferase – based reporter test [70]. It has also been observed in both B cell lymphoma and EBV – transformed lymphoblast cells, there is a direct interaction between EBNA3C and Gemim3. This interaction enhances the creation of p53 complexes with Gemim3 and hence reduces p53's DNA binding activity [71]. The overexpression of LMP1 has been found in EBV linked nasopharyngeal carcinomas, and it appears to accumulate with p53 through an unknown mechanism. Upon the activation of A20, LMP1 has been shown to block p53-mediated apoptosis [72][73]. In non-small-cell lung cancers when temperature sensitive p53 and LMP1 were stably expressed, causing LMP1 to prevent p53-mediated apoptosis through the promotion of the A20 gene expression [74]. The interaction of DNA damage with interferon regulatory factor 5 (IRF5) at the LMP1 promoter enhanced the endogenous production of LMP1 in EBV – transformed cells. Furthermore, ectopic IRF5 helps in boosting the endogenous LMP1 expression while inhibiting p53-mediated apoptosis [75]. BZLF1 (Z), an EBV immediate-early protein, interacts with p53 via its C-terminus region and inhibits p53-dependent transactivation in lymphoid cells, although the p53 overexpression can restores its function [76]. The formation of complexes between W repetitions of 66 amino acid long peptides of EBNA5 (EBNA-LP) and p53 has also been seen, however it is unknown how these complexes affect the progression of malignancies [77]

- **EBV & p63 ISOFORM INTERACTIONS:**

In human keratinocytes, a physical interaction of LMP2A with Np63 has been reported, resulting in enhanced expression and stabilization of Np63 in the cytoplasm and nuclear membrane. With the participation of PY and ITAM motifs, this connection is linked to calcium-induced disruption of cellular differentiation. LMP2A promoted Np63 expression through regulation of Itch over Np63, according to co – immunoprecipitation experiments [78]. Another co – immunoprecipitation study in nasopharyngeal carcinoma and EBV positive Burkitt's lymphoma described the direct connection between EBNA 5 and p63, which may contribute to the stability of p63, although the mechanism is yet unknown [79]. B cell lymphoma has been shown to interact with p63 and EBNA2. The amino acid sequence 310–336 in EBNA2 is recognised by P63 and binds to it. The GTGGGA motif loses recognition when the codon GTG> TCT is changed. GPPWWPP is the most prevalent motif observed in both type 1 and type 2 EBV EBNA2 sequences. The interaction of p63 is abolished when WW is changed to SS or FF, implying that WW's hydrophobic and aromatic properties are required for contact [80].

- **EBV & p73 ISOFORM INTERACTIONS:**

EBNA3C reduced the doxorubicin – induced apoptosis in p53-null cell lines (Saos-2 and HCT p53 double mutant) by downregulating the p73 protein production via stabilising Np73 [81]. In primary B cells infected with recombinant EBV, LMP1 has been shown to upregulate Np73. Chip tests revealed Np73 activation via p73 recruitment following the relocation of the poly-comb 2 complex component EZH2, as well as epigenetic alterations caused by c-Jun NH2-terminal kinase 1 activation (JNK-1), It was also seen that the Np73 expression levels were unaffected by the LMP-1 mutant lacking the JNK-1 activation domain (CTAR2) [82]. Single nucleotide polymorphisms (SNP's) in the p73 gene have been found to be crucial for the interaction with EBV in chronic lymphocytic leukaemia. Two p73 SNP's (rs3765701 and rs1885859) were shown to change the link between aberrant EBV and chronic lymphocytic leukaemia in the dominant model [83]. In EBV – related gastric cancer, DNA methylation in the CpG island of p73 was discovered. In comparison to EBV – negative gastric cancer, the immunohistochemistry analysis revealed a decrease of p73 expression in EBV – associated gastric cancer. In EBV – associated gastric cancer, a methylation-specific PCR reaction was used to reveal abnormal methylation patterns in exon 1 of the p73 gene [84].

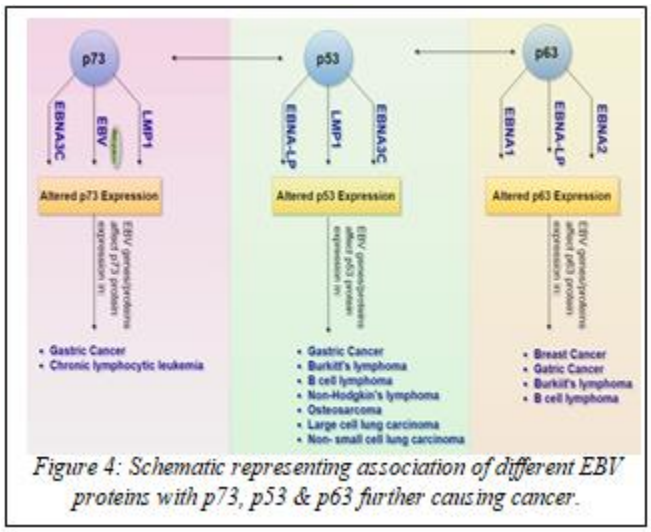

Figure 4: Schematic representing association of different EBV proteins with p73, p53 & p63 further causing cancer.

IV. EBV ASSOCIATED CANCERS

Since its identification as the first human tumour virus EBV has been linked to the development of a wide spectrum of malignancies. In order to be oncogenic EBV must maintain its viral genome in cell and prevent it from destroying. EBV establishes a latent infection in B cells to protect its viral genome. EBV gene products produced during lytic and latent phase interact with host genes, triggering dysregulation which may lead to malignancy [85]. The environmental and immunological components that contribute to the formation of these tumours are unclear, although they may be comparable to those that contribute to the development of EBV malignancies with endemic incidence patterns or related to immunological impairment. Environmental or genetic variables are likely to enhance infection of specific cell types and activate cellular pathways that are highly synergistic with EBV genes that regulate cell proliferation.

- **BURKITT'S LYMPHOMA:**

EBV was first revealed in cell lines derived from Burkitt's Lymphoma (BL), a rare paediatric cancer [86]. Burkitt's lymphomas a particularly aggressive lymphoma, the hallmark of which is a chromosomal translocation between chromosome 8 and either chromosomes 14, 2, or 22 [87]. The oncogene c-myc (chromosome 8) is juxtaposed to the immunoglobulin heavy-chain (chromosome 14) or light-chain genes as a result of this translocation (chromosomes 2 or 22). C-myc expression is deregulated as a result of this aberrant configuration [85]. It has been proposed that B-cell stimulation induced by continuous malaria reinfection contributes to an increased number of EBV-infected, proliferating B cells, which are more likely to carry cytogenetic abnormalities [88]. Only EBNA-1 and the EBER RNAs are expressed in EBV positive BL (Latency I) [89] Translocation of the c-myc gene to one of the immunoglobulin loci characterises all BL tumour cells, leading in altered c-myc regulation. Furthermore, mutations in the region of c-myc that typically binds the 107 repressors of myc function are frequently observed [90]. The BL type of lymphoma is observed in certain AIDS patients, and 50 % of them are EBV positive. Other oncogene alterations that have been found in BL are in pim- 1, c-fps/c-fes and ~53. P53 mutations have also been observed in around 30% of BL [37].

- **HODGKIN'S DISEASE:**

Hodgkin Lymphoma (HL) is a lymphatic malignancy distinguished by the presence of large, malignant lymphoid cells known as Reed-Sternberg cells. HL spreads between lymph node groups in a predictable and continuous manner [91]. In roughly 40% of HD patients, EBV is detected in the Reed-Sternberg cells, which are usually of the mixed cellularity type, and also are more frequently observed with males rather than females [92][92]. The involvement of EBV in Hodgkin's disease is currently under investigation. A latency II pattern of EBNA-1, LMP-1, LMP-2A, and LMP-2B, and EBERs have been observed [94][95]. Evidence suggests that the prevalence of EBV-positive Hodgkin's disease is age-related, with the virus being more commonly associated with malignancies in children and the elderly [96]. The prevalence of EBV positive in young children may be due to primary EBV infection, the relationship of EBV with tumour in older individuals may be due to enhanced viral activity as a result of flagging T-cells immunity [91].

- **NASOPHARYNGEAL CARCINOMA:**

Nasopharyngeal carcinoma (NPC) is a malignant epithelial tumour that affects the mucosa of the nasopharynx, which is rich in lymphocytes. Multiple etiological variables, such as genetic predisposition, nutrition, and Epstein-Barr virus (EBV) infection, influence the pathogenesis of this form of head and neck cancer [97][97]. Although no EBV-compatible receptor has been discovered on epithelial cells, a surface protein antigenically similar to the B cell has been reported. EBV might potentially exploit the CD21 receptor as a point of entry [99]. Alternatively, EBV could also enter nasopharyngeal cells via IgA-mediated endocytosis [100]. In Nasopharyngeal Cancer EBV genes expression includes EBNA-1 (from Qp) with the EBER RNAs, LMP-1 (from Qp) with the EBER RNAs, LMP- 2A, B and (in about 65% of cases), they undergo latency II pattern. BamHI A region transcripts (BARFO, CSTs) have also been reported [101]. P16 Cyclin dependent kinase inhibitor gene (9q21-22) is mostly found altered. NPC [102][103]. Changes in other chromosomal regions have also been also observed such as 3p13-14.3, 11q13.3-22 and 11q22-24, although no affected genes have been observed in this region [104][104]. P53 mutations have also been observed in few NPC cases. Bcl-2 overexpression may potentially contribute to oncogenesis by enabling cells to avoid apoptosis [106][106]. Despite the fact that nasopharyngeal cancer cells have normal antigen processing and are recognised by EBV-specific CTLs, still they are not killed [108]. Increased production of IL-1 and IL-1 by epithelial cells and CD4 T cells has been linked to increased production of EBV-encoded viral IL-10 in nasopharyngeal carcinoma, which may contribute to tumour progression and immune evasion [109].

- **GASTRIC CANCER:**

When cancer cells develop in the lining of the stomach, it is known as gastric cancer. EBV-positive gastric cancer has distinct genetic abnormalities, substantial clinicopathological

characteristics, and a favourable prognosis. After EBV infects the human body, it goes through an incubation phase during which the virus integrates its DNA into the host and expresses the latent protein. The latent protein subsequently alters DNA methylation through mi-RNA, resulting in EBV-positive gastric cancer [110]. EBV-positive gastric cancers are more likely in males compared with females EBERs, EBNA-1, miR-BARTS, and LMP2A are abundantly expressed and play an essential role in viral replication in EBV-positive gastric cancer, which is type I latency [111][112]. For EBV latent infection, EBNA-1 is a crucial molecule [113][113]. It promotes EBV adhesion to the host cell chromosome by binding to the viral oriP sequence in a sequence-dependent way [115]. EBNA-1 can also cause the loss of promyelocytic leukaemia protein nuclear bodies in the nucleosome and impair the cellular response to DNA damage in promyelocytic leukaemia [116]. In the latent condition of EBV infection, BARF-0 and BARF-1 are also essential [117]. It can also affect the NF-B/miR-146a/Smad4 pathway and cyclin-D1 protein expression in gastric cancer cells [118]. Furthermore, BARF-1 has the ability to activate the cell cycle regulator bcl-2 [119]. Gastric cancer cells proliferate as a result of these mechanisms.

V. THERAPEUTIC APPROACHES

- **CRISPR BASED THERAPEUTIC APPROACH FOR TREATMENT OF EBV – ASSOCIATED LYMPHOMAS:**

The CRISPR/Cas9 system's development offers a potential technique for ending latent infection. The CRISPR/Cas9 system works by using guide RNAs (gRNAs) to target the EBNA1 and oriP regions. With this technique a more than 95% loss of EBV genomes was reported [120]. The CRISPR/Cas9 screen has also been used to uncover important variables that affect viral latency or the lytic programme, as well as to investigate possible treatment targets for EBV – associated lymphomas. Through a CRISPR/Cas9 system screen, it was discovered that the ubiquitin ligase ubiquitin-like PHD and RING finger domain-containing protein 1 (UHRF1), as well as DNA methyl-transferases (DNMT1 and DNMT3B), which found to be as potent oncoproteins in EBV – latency III linked to Burkitt's Lymphoma [121]. Histone ubiquitination mediated by the Polycomb Repressive Complex I (PRC1) is another way for limiting viral latency. These findings add to the list of reasonable and important therapeutic targets for viral protein expression regulation [121].

- **THERAPY BASED ON CHIMERIC ANTIGEN RECEPTOR T – CELL:**

T-cells using Chimeric Antigen Receptors (CARs) are designed to circumvent the resistance that exists in traditional adoptive therapy for tumours that lack MHC antigen manifestation system [122]. CD19-specific CAR T-cells are presently the best-studied CAR T-cells for treating B-cell malignancies. CD19 is a great target because it's only found on B-cells. Although CD19 CAR T-cells have shown to be effective, but they are also linked to some of the side effects such as cytokine release syndrome, B-cell aplasia, encephalopathy & off-target B-cell mortality [123]. Latent membrane proteins, which are present on the cell surface during latency programmes II and III, are a potential CAR target in EBV – associated lymphoma. An infusion of LMP1 HELA/CAR T-cells suppressed NPC tumour growth in a xenograft model, indicating that this could be a potential strategy against nasopharyngeal cancer in vitro and in a mouse model. This strategy could work in LMP1-positive lymphomas as well, although more research is needed [124].

- **MONOCLONAL ANTIBODY BASED THERAPY:**

Rituximab, an anti-CD20 monoclonal antibody, has shown promising results in the treatment of a variety of CD20-expressing lymphomas. It is also effective in the treatment of lymphoproliferative diseases caused by EBV. It is currently being used to treat Post-Transplant Lymphoproliferative Disorder (PTLD) by inducing depletion of B-cell, and the reported overall response rate to anti-CD20 treatment alone ranges from 32% to 79% in PTLD patients [125]. Rituximab is also administered in EBV-positive patients in conjunction with cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) [126][126]. Overall, the toxicity associated with rituximab has been shown to be relatively tolerable. Recent research has investigated at the idea of utilising rituximab as a prophylactic for patients with suspected EBV reactivation identified in their peripheral blood, either pre-transplant or post-transplant. Pre-transplant rituximab decreased the overall incidence of PTLD, and most patients who received pre-emptive rituximab post-transplant did not develop PTLD [127][127]. Although anti-CD20 has been shown to be beneficial in EBV-positive lymphomas, CD20 is expressed on most normal B cells, therefore it does not exclusively target EBV-positive cells. CD70, which is exclusively expressed in highly activated B- or T-cells, has been proposed as a potential target in EBV-associated lymphomas [129].

- **EBV VACCINE:**

Despite the difficulty in developing a live vaccine, work on component EBV vaccines for preventative and therapeutic purposes has continued. Most vaccines employ the gp350 glycoprotein as an antigen. It is the principal antigen for inducing neutralizing antibodies against B-cell infection in human sera [130][131]. EBV gp350 mediates B-cell infection by binding to the complement receptor 2 (CR2/CD21) and it represents a promising target for neutralizing antibodies. Antigens such as EBNA1 and LMP2A have also been utilised [131]. Another method is being studied in addition to the creation of component vaccinations. AMMOM1 is an EBV gH/gL monoclonal antibody that prevents viral infection of B cells and epithelial cells [132]. It may be possible to produce a novel form of EBV vaccination by using the epitope identified by the antibody. mRNA vaccines also represent promising approaches. Currently mRNA vaccine is being developed encoding five EBV glycoproteins (gp350, gH/gL/gp42, and gB) by Moderna Therapeutics, it may reduce the rate of EBV-associated infectious mononucleosis (IM) and possibly prevent EBV infection. As this mRNA vaccine is in preclinical development, so the efficacy still needs further investigation. Furthermore, molecular Clamp is a novel technique that produces chimeric polypeptides that imitate

the pre-fusion structures of viral fusion proteins, allowing for the fast development of anti-fusion vaccines or inhibitors against enveloped viruses [133]. This innovative approach has also been utilised to create vaccines for influenza, HIV, Ebola, and SARS-CoV-2.

VI. CONCLUSION

Here, we discussed EBV infection, Role of EBV in Malignancies, EBV viral genome expression, Interaction of EBV with P53 family as well as clinical and therapeutic approach in EBV infection. EBV mainly known for causing mononucleosis is largely associated with mutation of P53 family thereby preventing cell apoptosis. EBV initially infects the oropharyngeal epithelial cell, it enters the circulating B lymphocyte, and remains in a dormant condition for the rest of one's life. During its lytic and latent phase several EBV gene products are secreted which interacts with P53, P63 and P73. Despite structural and functional similarities, there are several functional variations between p53, P63, and p73 in terms of cell cycle regulation and cancer which are briefly described. The capability of various tumour virus gene products to bind and inactivate p53, thereby blocking its function in mediating growth arrest or apoptosis, has defined a role for both p53 and viral oncogenes in the cell transformation pathway. EBV has been associated to the development of a wide range of cancers since its discovery as the first human malignant virus. EBV infection is closely linked to Burkitt's lymphoma, Hodgkin lymphoma, Nasopharyngeal carcinoma (NPC), Gastric cancers as well as its relation with Breast cancer is also established. It is yet unclear whether EBV altering the expression of one of the p53 family proteins regulates the expression of other p53 family proteins in the same array or in a different way. To learn more about the processes behind EBV-induced p53/p63/p73 regulation in EBV-associated malignancies in specific tissues, additional study is essential. Therapy for EBV-associated tumours remains largely in the nascent stages, but research is being fuelled for successful development of new therapeutic and immunological approaches.

REFERNECES

[1]. G. Niedobitek, N. Meru, and H. J. Delecluse, "Epstein-Barr virus infection and human malignancies," *Int J Exp Pathol*, vol. 82, no. 3, pp. 149–170, Jun. 2001, doi: 10.1046/j.1365-2613.2001.iep0082-0149-x.

[2]. R. J. Kraus *et al.*, "Reactivation of Epstein-Barr Virus by HIF-1α Requires p53," *J Virol*, vol. 94, no. 18, Aug. 2020, doi: 10.1128/JVI.00722-20.

[3]. M. K. Smatti, D. W. Al-Sadeq, N. H. Ali, G. Pintus, H. Abou-Saleh, and G. K. Nasrallah, "Epstein–Barr Virus Epidemiology, Serology, and Genetic Variability of LMP-1 Oncogene Among Healthy Population: An Update," *Front. Oncol.*, vol. 8, p. 211, Jun. 2018, doi: 10.3389/fonc.2018.00211.

[4]. J. Ribeiro *et al.*, "Clinical and pathological characterization of Epstein-Barr virus-associated gastric carcinomas in Portugal," *World J Gastroenterol*, vol. 23, no. 40, pp. 7292–7302, Oct. 2017, doi: 10.3748/wjg.v23.i40.7292.

[5]. J. Ribeiro *et al.*, "P53 deregulation in Epstein-Barr virus-associated gastric cancer," *Cancer Letters*, vol. 404, pp. 37–43, Sep. 2017, doi: 10.1016/j.canlet.2017.07.010.

[6]. M.-S. Kang and E. Kieff, "Epstein–Barr virus latent genes," *Exp Mol Med*, vol. 47, no. 1, Art. no. 1, Jan. 2015, doi: 10.1038/emm.2014.84.

[7]. O. Indari, R. Chandramohanadas, and H. C. Jha, "Epstein–Barr virus infection modulates blood–brain barrier cells and its co-infection with *Plasmodium falciparum* induces RBC adhesion," *Pathogens and Disease*, vol. 79, no. 1, p. ftaa080, Jan. 2021, doi: 10.1093/femspd/ftaa080.

[8]. B. Leroy, M. Anderson, and T. Soussi, "TP53 mutations in human cancer: database reassessment and prospects for the next decade," *Hum Mutat*, vol. 35, no. 6, pp. 672–688, Jun. 2014, doi: 10.1002/humu.22552.

[9]. K. Chatterjee, P. Das, N. R. Chattopadhyay, S. Mal, and T. Choudhuri, "The interplay between Epstein-Bar virus (EBV) with the p53 and its homologs during EBV associated malignancies," *Heliyon*, vol. 5, no. 11, p. e02624, Nov. 2019, doi: 10.1016/j.heliyon.2019.e02624.

[10]. S. Murono, T. Yoshizaki, C. S. Park, and M. Furukawa, "Association of Epstein-Barr virus infection with p53 protein accumulation but not bcl-2 protein in nasopharyngeal carcinoma," *Histopathology*, vol. 34, no. 5, pp. 432–438, May 1999, doi: 10.1046/j.1365-2559.1999.00625.x.

[11]. A. J. Bass *et al.*, "Comprehensive molecular characterization of gastric adenocarcinoma," *Nature*, vol. 513, no. 7517, pp. 202–209, Sep. 2014, doi: 10.1038/nature13480.

[12]. S. D. Hudnall, "Epstein–Barr Virus: Pathogenesis and Host Immune Response," in *Viruses and Human Cancer*, S. D. Hudnall, Ed. New York, NY: Springer New York, 2014, pp. 7–24. doi: 10.1007/978-1-4939-0870-7_2.

[13]. D. H. Crawford *et al.*, "A Cohort Study among University Students: Identification of Risk Factors for Epstein-Barr Virus Seroconversion and Infectious Mononucleosis," *CLIN INFECT DIS*, vol. 43, no. 3, pp. 276–282, Aug. 2006, doi: 10.1086/505400.

[14]. J. C. Niederman, R. W. McCollum, G. Henle, and W. Henle, "Infectious mononucleosis. Clinical manifestations in relation to EB virus antibodies," *JAMA*, vol. 203, no. 3, pp. 205–209, Jan. 1968, doi: 10.1001/jama.203.3.205.

[15]. D. A. Thorley-Lawson and A. Gross, "Persistence of the Epstein-Barr virus and the origins of associated lymphomas," *N Engl J Med*, vol. 350, no. 13, pp. 1328–1337, Mar. 2004, doi: 10.1056/NEJMra032015.

[16]. J. W. Sixbey, P. Shirley, P. J. Chesney, D. M. Buntin, and L. Resnick, "Detection of a second widespread strain of Epstein-Barr virus," *Lancet*, vol. 2, no. 8666, pp. 761–765, Sep. 1989, doi: 10.1016/s0140-6736(89)90829-5.

[17]. A. Apolloni and T. B. Sculley, "Detection of A-type and B-type Epstein-Barr virus in throat washings and lymphocytes," *Virology*, vol. 202, no. 2, pp. 978–981, Aug. 1994, doi: 10.1006/viro.1994.1422.
[18]. D. M. Walling, C. M. Flaitz, C. M. Nichols, S. D. Hudnall, and K. Adler-Storthz, "Persistent productive Epstein-Barr virus replication in normal epithelial cells in vivo," *J Infect Dis*, vol. 184, no. 12, pp. 1499–1507, Dec. 2001, doi: 10.1086/323992.
[19]. K. Herrmann, P. Frangou, J. Middeldorp, and G. Niedobitek, "Epstein-Barr virus replication in tongue epithelial cells," *J Gen Virol*, vol. 83, no. Pt 12, pp. 2995–2998, Dec. 2002, doi: 10.1099/0022-1317-83-12-2995.
[20]. P. Frangou, M. Buettner, and G. Niedobitek, "Epstein-Barr virus (EBV) infection in epithelial cells in vivo: rare detection of EBV replication in tongue mucosa but not in salivary glands," *J Infect Dis*, vol. 191, no. 2, pp. 238–242, Jan. 2005, doi: 10.1086/426823.
[21]. M. Kalla, A. Schmeinck, M. Bergbauer, D. Pich, and W. Hammerschmidt, "AP-1 homolog BZLF1 of Epstein–Barr virus has two essential functions dependent on the epigenetic state of the viral genome," *PNAS*, vol. 107, no. 2, pp. 850–855, Jan. 2010, doi: 10.1073/pnas.0911948107.
[22]. E. Seto, A. Moosmann, S. Grömminger, N. Walz, A. Grundhoff, and W. Hammerschmidt, "Micro RNAs of Epstein-Barr Virus Promote Cell Cycle Progression and Prevent Apoptosis of Primary Human B Cells," *PLOS Pathogens*, vol. 6, no. 8, p. e1001063, Aug. 2010, doi: 10.1371/journal.ppat.1001063.
[23]. R. J. Tierney, N. Steven, L. S. Young, and A. B. Rickinson, "Epstein-Barr virus latency in blood mononuclear cells: analysis of viral gene transcription during primary infection and in the carrier state," *J Virol*, vol. 68, no. 11, pp. 7374–7385, Nov. 1994, doi: 10.1128/JVI.68.11.7374-7385.1994.
[24]. A. D. Hislop, G. S. Taylor, D. Sauce, and A. B. Rickinson, "Cellular responses to viral infection in humans: lessons from Epstein-Barr virus," *Annu Rev Immunol*, vol. 25, pp. 587–617, 2007, doi: 10.1146/annurev.immunol.25.022106.141553.
[25]. M. De Paschale and P. Clerici, "Serological diagnosis of Epstein-Barr virus infection: Problems and solutions," *World J Virol*, vol. 1, no. 1, pp. 31–43, Feb. 2012, doi: 10.5501/wjv.v1.i1.31.
[26]. O. Chijioke, T. Azzi, D. Nadal, and C. Münz, "Innate immune responses against Epstein Barr virus infection," *J Leukoc Biol*, vol. 94, no. 6, pp. 1185–1190, Dec. 2013, doi: 10.1189/jlb.0313173.
[27]. J. L. Yates, N. Warren, and B. Sugden, "Stable replication of plasmids derived from Epstein-Barr virus in various mammalian cells," *Nature*, vol. 313, no. 6005, pp. 812–815, Mar. 1985, doi: 10.1038/313812a0.
[28]. R. G. Caldwell, J. B. Wilson, S. J. Anderson, and R. Longnecker, "Epstein-Barr virus LMP2A drives B cell development and survival in the absence of normal B cell receptor signals," *Immunity*, vol. 9, no. 3, pp. 405–411, Sep. 1998, doi: 10.1016/s1074-7613(00)80623-8.
[29]. O. Gires *et al.*, "Latent membrane protein 1 of Epstein–Barr virus interacts with JAK3 and activates STAT proteins," *The EMBO Journal*, vol. 18, no. 11, pp. 3064–3073, Jun. 1999, doi: 10.1093/emboj/18.11.3064.
[30]. G. J. Babcock, L. L. Decker, M. Volk, and D. A. Thorley-Lawson, "EBV persistence in memory B cells in vivo," *Immunity*, vol. 9, no. 3, pp. 395–404, Sep. 1998, doi: 10.1016/s1074-7613(00)80622-6.
[31]. G. J. Babcock and D. A. Thorley-Lawson, "Tonsillar memory B cells, latently infected with Epstein–Barr virus, express the restricted pattern of latent genes previously found only in Epstein–Barr virus-associated tumors," *Proc Natl Acad Sci U S A*, vol. 97, no. 22, pp. 12250–12255, Oct. 2000.
[32]. D. A. Thorley-Lawson and G. J. Babcock, "A model for persistent infection with Epstein-Barr virus: the stealth virus of human B cells," *Life Sci*, vol. 65, no. 14, pp. 1433–1453, 1999, doi: 10.1016/s0024-3205(99)00214-3.
[33]. D. Hochberg, J. M. Middeldorp, M. Catalina, J. L. Sullivan, K. Luzuriaga, and D. A. Thorley-Lawson, "Demonstration of the Burkitt's lymphoma Epstein-Barr virus phenotype in dividing latently infected memory cells in vivo," *Proc Natl Acad Sci U S A*, vol. 101, no. 1, pp. 239–244, Jan. 2004, doi: 10.1073/pnas.2237267100.
[34]. I. Anagnostopoulos, M. Hummel, C. Kreschel, and H. Stein, "Morphology, immunophenotype, and distribution of latently and/or productively Epstein-Barr virus-infected cells in acute infectious mononucleosis: implications for the interindividual infection route of Epstein-Barr virus," *Blood*, vol. 85, no. 3, pp. 744–750, Feb. 1995.
[35]. L. L. Laichalk and D. A. Thorley-Lawson, "Terminal differentiation into plasma cells initiates the replicative cycle of Epstein-Barr virus in vivo," *J Virol*, vol. 79, no. 2, pp. 1296–1307, Jan. 2005, doi: 10.1128/JVI.79.2.1296-1307.2005.
[36]. L. C. Tan *et al.*, "A re-evaluation of the frequency of CD8+ T cells specific for EBV in healthy virus carriers," *J Immunol*, vol. 162, no. 3, pp. 1827–1835, Feb. 1999.
[37]. P. J. Farrell, I. Cludts, and A. Stühler, "Epstein-Barr virus genes and cancer cells," *Biomed Pharmacother*, vol. 51, no. 6–7, pp. 258–267, 1997, doi: 10.1016/s0753-3322(97)83541-x.
[38]. T. A. Gahn and B. Sugden, "An EBNA-1-dependent enhancer acts from a distance of 10 kilobase pairs to increase expression of the Epstein-Barr virus LMP gene," *J Virol*, vol. 69, no. 4, pp. 2633–2636, Apr. 1995, doi: 10.1128/JVI.69.4.2633-2636.1995.
[39]. R. F. Ambinder, W. A. Shah, D. R. Rawlins, G. S. Hayward, and S. D. Hayward, "Definition of the sequence requirements for binding of the EBNA-1 protein to its palindromic target sites in Epstein-Barr virus DNA," *J Virol*, vol. 64, no. 5, pp. 2369–2379, May 1990, doi: 10.1128/JVI.64.5.2369-2379.1990.
[40]. C. H. Jones, S. D. Hayward, and D. R. Rawlins, "Interaction of the lymphocyte-derived Epstein-Barr virus nuclear antigen EBNA-1 with its DNA-binding

sites," *J Virol*, vol. 63, no. 1, pp. 101–110, Jan. 1989, doi: 10.1128/JVI.63.1.101-110.1989.

[41]. D. R. Rawlins, G. Milman, S. D. Hayward, and G. S. Hayward, "Sequence-specific DNA binding of the Epstein-Barr virus nuclear antigen (EBNA-1) to clustered sites in the plasmid maintenance region," *Cell*, vol. 42, no. 3, pp. 859–868, Oct. 1985, doi: 10.1016/0092-8674(85)90282-x.

[42]. D. A. Wysokenski and J. L. Yates, "Multiple EBNA1-binding sites are required to form an EBNA1-dependent enhancer and to activate a minimal replicative origin within oriP of Epstein-Barr virus," *J Virol*, vol. 63, no. 6, pp. 2657–2666, Jun. 1989, doi: 10.1128/JVI.63.6.2657-2666.1989.

[43]. B. Wensing and P. J. Farrell, "Regulation of cell growth and death by Epstein-Barr virus," *Microbes Infect*, vol. 2, no. 1, pp. 77–84, Jan. 2000, doi: 10.1016/s1286-4579(00)00282-3.

[44]. J. M. Lee, K.-H. Lee, M. Weidner, B. A. Osborne, and S. D. Hayward, "Epstein-Barr virus EBNA2 blocks Nur77- mediated apoptosis," *Proc Natl Acad Sci U S A*, vol. 99, no. 18, pp. 11878–11883, Sep. 2002, doi: 10.1073/pnas.182552499.

[45]. S. A. Radkov, M. Bain, P. J. Farrell, M. West, M. Rowe, and M. J. Allday, "Epstein-Barr virus EBNA3C represses Cp, the major promoter for EBNA expression, but has no effect on the promoter of the cell gene CD21," *J Virol*, vol. 71, no. 11, pp. 8552–8562, Nov. 1997, doi: 10.1128/JVI.71.11.8552-8562.1997.

[46]. C. D. Wood *et al.*, "MYC activation and BCL2L11 silencing by a tumour virus through the large-scale reconfiguration of enhancer-promoter hubs," *Elife*, vol. 5, Aug. 2016, doi: 10.7554/eLife.18270.

[47]. A. J. Sinclair, I. Palmero, G. Peters, and P. J. Farrell, "EBNA-2 and EBNA-LP cooperate to cause G0 to G1 transition during immortalization of resting human B lymphocytes by Epstein-Barr virus," *EMBO J*, vol. 13, no. 14, pp. 3321–3328, Jul. 1994.

[48]. P. G. Murray and L. S. Young, "The Role of the Epstein-Barr virus in human disease," *Front Biosci*, vol. 7, pp. d519-540, Feb. 2002, doi: 10.2741/murray.

[49]. B. Tomkinson and E. Kieff, "Use of second-site homologous recombination to demonstrate that Epstein-Barr virus nuclear protein 3B is not important for lymphocyte infection or growth transformation in vitro," *J Virol*, vol. 66, no. 5, pp. 2893–2903, May 1992, doi: 10.1128/JVI.66.5.2893-2903.1992.

[50]. G. A. Parker, T. Crook, M. Bain, E. A. Sara, P. J. Farrell, and M. J. Allday, "Epstein-Barr virus nuclear antigen (EBNA)3C is an immortalizing oncoprotein with similar properties to adenovirus E1A and papillomavirus E7," *Oncogene*, vol. 13, no. 12, pp. 2541–2549, Dec. 1996.

[51]. M. J. Allday and P. J. Farrell, "Epstein-Barr virus nuclear antigen EBNA3C/6 expression maintains the level of latent membrane protein 1 in G1-arrested cells," *J Virol*, vol. 68, no. 6, pp. 3491–3498, Jun. 1994, doi: 10.1128/JVI.68.6.3491-3498.1994.

[52]. S. L. Silins and T. B. Sculley, "Modulation of vimentin, the CD40 activation antigen and Burkitt's lymphoma antigen (CD77) by the Epstein-Barr virus nuclear antigen EBNA-4," *Virology*, vol. 202, no. 1, pp. 16–24, Jul. 1994, doi: 10.1006/viro.1994.1317.

[53]. A. Joutel and E. Tournier-Lasserve, "Notch signalling pathway and human diseases," *Semin Cell Dev Biol*, vol. 9, no. 6, pp. 619–625, Dec. 1998, doi: 10.1006/scdb.1998.0261.

[54]. O. Gires *et al.*, "Latent membrane protein 1 of Epstein-Barr virus mimics a constitutively active receptor molecule.," *EMBO J*, vol. 16, no. 20, pp. 6131–6140, Oct. 1997, doi: 10.1093/emboj/16.20.6131.

[55]. L. W. Wang, S. Jiang, and B. E. Gewurz, "Epstein-Barr Virus LMP1-Mediated Oncogenicity," *J Virol*, vol. 91, no. 21, Nov. 2017, doi: 10.1128/JVI.01718-16.

[56]. L. S. Young and A. B. Rickinson, "Epstein–Barr virus: 40 years on," *Nat Rev Cancer*, vol. 4, no. 10, pp. 757–768, Oct. 2004, doi: 10.1038/nrc1452.

[57]. J. Pflaum, S. Schlosser, and M. Müller, "p53 Family and Cellular Stress Responses in Cancer," *Front Oncol*, vol. 4, p. 285, 2014, doi: 10.3389/fonc.2014.00285.

[58]. K. D. Sullivan, M. D. Galbraith, Z. Andrysik, and J. M. Espinosa, "Mechanisms of transcriptional regulation by p53," *Cell Death Differ*, vol. 25, no. 1, pp. 133–143, Jan. 2018, doi: 10.1038/cdd.2017.174.

[59]. N. C. for B. Information (US), *The p53 tumor suppressor protein*. National Center for Biotechnology Information (US), 1998. Accessed: Jun. 12, 2021. [Online]. Available: https://www.ncbi.nlm.nih.gov/books/NBK22268/

[60]. F. Murray-Zmijewski, D. P. Lane, and J.-C. Bourdon, "p53/p63/p73 isoforms: an orchestra of isoforms to harmonise cell differentiation and response to stress," *Cell Death Differ*, vol. 13, no. 6, pp. 962–972, Jun. 2006, doi: 10.1038/sj.cdd.4401914.

[61]. E. Candi, M. Agostini, G. Melino, and F. Bernassola, "How the TP53 family proteins TP63 and TP73 contribute to tumorigenesis: regulators and effectors," *Hum Mutat*, vol. 35, no. 6, pp. 702–714, Jun. 2014, doi: 10.1002/humu.22523.

[62]. A. Yang *et al.*, "p63 is essential for regenerative proliferation in limb, craniofacial and epithelial development," *Nature*, vol. 398, no. 6729, pp. 714–718, Apr. 1999, doi: 10.1038/19539.

[63]. A. Yang *et al.*, "p73-deficient mice have neurological, pheromonal and inflammatory defects but lack spontaneous tumours," *Nature*, vol. 404, no. 6773, pp. 99–103, Mar. 2000, doi: 10.1038/35003607.

[64]. U. M. Moll and N. Slade, "p63 and p73: roles in development and tumor formation," *Mol Cancer Res*, vol. 2, no. 7, pp. 371–386, Jul. 2004.

[65]. J. Wei, E. Zaika, and A. Zaika, "p53 Family: Role of Protein Isoforms in Human Cancer," *J Nucleic Acids*, vol. 2012, p. 687359, 2012, doi: 10.1155/2012/687359.

[66]. E. Soares and H. Zhou, "Master regulatory role of p63 in epidermal development and disease," *Cell Mol Life Sci*, vol. 75, no. 7, pp. 1179–1190, Apr. 2018, doi: 10.1007/s00018-017-2701-z.

[67]. G. Melino, X. Lu, M. Gasco, T. Crook, and R. A. Knight, "Functional regulation of p73 and p63: development and cancer," *Trends in Biochemical Sciences*, vol. 28, no. 12, pp. 663–670, Dec. 2003, doi: 10.1016/j.tibs.2003.10.004.

[68]. S. S. Lok, J. P. Stewart, B. G. Kelly, P. S. Hasleton, and J. J. Egan, "Epstein–Barr virus and wild p53 in idiopathic pulmonary fibrosis," *Respiratory Medicine*, vol. 95, no. 10, pp. 787–791, Oct. 2001, doi: 10.1053/rmed.2001.1152.

[69]. S. K. Shukla, H. C. Jha, D. W. El-Naccache, and E. S. Robertson, "An EBV recombinant deleted for residues 130-159 in EBNA3C can deregulate p53/Mdm2 and Cyclin D1/CDK6 which results in apoptosis and reduced cell proliferation," *Oncotarget*, vol. 7, no. 14, pp. 18116–18134, Feb. 2016, doi: 10.18632/oncotarget.7502.

[70]. F. Yi *et al.*, "Epstein–Barr virus nuclear antigen 3C targets p53 and modulates its transcriptional and apoptotic activities," *Virology*, vol. 388, no. 2, pp. 236–247, Jun. 2009, doi: 10.1016/j.virol.2009.03.027.

[71]. Q. Cai *et al.*, "Epstein-Barr Virus Nuclear Antigen 3C Stabilizes Gemin3 to Block p53-mediated Apoptosis," *PLOS Pathogens*, vol. 7, no. 12, p. e1002418, Dec. 2011, doi: 10.1371/journal.ppat.1002418.

[72]. M.-T. Liu *et al.*, "Epstein–Barr virus latent membrane protein 1 represses p53-mediated DNA repair and transcriptional activity," *Oncogene*, vol. 24, no. 16, Art. no. 16, Apr. 2005, doi: 10.1038/sj.onc.1208319.

[73]. J.-Y. Shao, I. Ernberg, P. Biberfeld, T. Heiden, Y.-X. Zeng, and L.-F. Hu, "Epstein-Barr Virus LMP1 Status in Relation to Apoptosis, P53 Expression and Leucocyte Infiltration in Nasopharyngeal Carcinoma," *ANTICANCER RESEARCH*, p. 10, 2004.

[74]. K. L. Fries, W. E. Miller, and N. Raab-Traub, "Epstein-Barr virus latent membrane protein 1 blocks p53-mediated apoptosis through the induction of the A20 gene," *J Virol*, vol. 70, no. 12, pp. 8653–8659, Dec. 1996, doi: 10.1128/jvi.70.12.8653-8659.1996.

[75]. Q. Wang, A. Lingel, V. Geiser, Z. Kwapnoski, and L. Zhang, "Tumor Suppressor p53 Stimulates the Expression of Epstein-Barr Virus Latent Membrane Protein 1," *J Virol*, vol. 91, no. 20, Oct. 2017, doi: 10.1128/JVI.00312-17.

[76]. Q. Zhang, D. Gutsch, and S. Kenney, "Functional and physical interaction between p53 and BZLF1: implications for Epstein-Barr virus latency.," *Mol Cell Biol*, vol. 14, no. 3, pp. 1929–1938, Mar. 1994.

[77]. L. Szekely, G. Selivanova, K. P. Magnusson, G. Klein, and K. G. Wiman, "EBNA-5, an Epstein-Barr virus-encoded nuclear antigen, binds to the retinoblastoma and p53 proteins," *Proc Natl Acad Sci U S A*, vol. 90, no. 12, pp. 5455–5459, Jun. 1993, doi: 10.1073/pnas.90.12.5455.

[78]. J. A. Fotheringham, S. Mazzucca, and N. Raab-Traub, "Epstein-Barr virus latent membrane protein-2A-induced ΔNp63α expression is associated with impaired epithelial-cell differentiation," *Oncogene*, vol. 29, no. 30, Art. no. 30, Jul. 2010, doi: 10.1038/onc.2010.175.

[79]. C. Guo *et al.*, "The expression of p63 is associated with the differential stage in nasopharyngeal carcinoma and EBV infection," *J Transl Med*, vol. 4, p. 23, May 2006, doi: 10.1186/1479-5876-4-23.

[80]. R. Yalamanchili, X. Tong, S. Grossman, E. Johannsen, G. Mosialos, and E. Kieff, "Genetic and biochemical evidence that EBNA 2 interaction with a 63-kDa cellular GTG-binding protein is essential for B lymphocyte growth transformation by EBV," *Virology*, vol. 204, no. 2, pp. 634–641, Nov. 1994, doi: 10.1006/viro.1994.1578.

[81]. S. K. Sahu, S. Mohanty, A. Kumar, C. N. Kundu, S. C. Verma, and T. Choudhuri, "Epstein-Barr virus nuclear antigen 3C interact with p73: Interplay between a viral oncoprotein and cellular tumor suppressor," *Virology*, vol. 448, pp. 333–343, Jan. 2014, doi: 10.1016/j.virol.2013.10.023.

[82]. R. Accardi *et al.*, "Epstein - Barr Virus Transforming Protein LMP-1 Alters B Cells Gene Expression by Promoting Accumulation of the Oncoprotein ΔNp73α," *PLOS Pathogens*, vol. 9, no. 3, p. e1003186, Mar. 2013, doi: 10.1371/journal.ppat.1003186.

[83]. D. Casabonne *et al.*, "Single nucleotide polymorphisms of matrix metalloproteinase 9 (MMP9) and tumor protein 73 (TP73) interact with Epstein-Barr virus in chronic lymphocytic leukemia: results from the European case-control study EpiLymph," *HAEMATOL-HEMATOL J*, vol. 96, no. 2, pp. 323–327, 2011, doi: 10.3324/haematol.2010.031161.

[84]. T. Ushiku *et al.*, "p73 gene promoter methylation in Epstein-Barr virus-associated gastric carcinoma," *International Journal of Cancer*, vol. 120, no. 1, pp. 60–66, 2007, doi: 10.1002/ijc.22275.

[85]. M. P. Thompson and R. Kurzrock, "Epstein-Barr Virus and Cancer," p. 20.

[86]. M. A. Epstein, B. G. Achong, and Y. M. Barr, "VIRUS PARTICLES IN CULTURED LYMPHOBLASTS FROM BURKITT'S LYMPHOMA," *Lancet*, vol. 1, no. 7335, pp. 702–703, Mar. 1964, doi: 10.1016/s0140-6736(64)91524-7.

[87]. R. Taub *et al.*, "Translocation of the c-myc gene into the immunoglobulin heavy chain locus in human Burkitt lymphoma and murine plasmacytoma cells," *Proc Natl Acad Sci U S A*, vol. 79, no. 24, pp. 7837–7841, Dec. 1982, doi: 10.1073/pnas.79.24.7837.

[88]. S. F. Lyons and D. N. Liebowitz, "The roles of human viruses in the pathogenesis of lymphoma," *Semin Oncol*, vol. 25, no. 4, pp. 461–475, Aug. 1998.

[89]. M. Rowe *et al.*, "Differences in B cell growth phenotype reflect novel patterns of Epstein-Barr virus latent gene expression in Burkitt's lymphoma cells," *EMBO J*, vol. 6, no. 9, pp. 2743–2751, Sep. 1987.

[90]. W. Gu, K. Bhatia, I. T. Magrath, C. V. Dang, and R. Dalla-Favera, "Binding and suppression of the Myc transcriptional activation domain by p107," *Science*, vol. 264, no. 5156, pp. 251–254, Apr. 1994, doi: 10.1126/science.8146655.

[91]. A. Maggioncalda, N. Malik, P. Shenoy, M. Smith, R. Sinha, and C. R. Flowers, "Clinical, Molecular, and Environmental Risk Factors for Hodgkin Lymphoma," *Advances in Hematology*, vol. 2011, p. e736261, Nov. 2010, doi: 10.1155/2011/736261.

[92]. Y. Tomita *et al.*, "Epstein-Barr virus in Hodgkin's disease patients in Japan," *Cancer*, vol. 77, no. 1, pp. 186–192, 1996, doi: 10.1002/(SICI)1097-0142(19960101)77:1<186::AID-CNCR30>3.0.CO;2-#.

[93]. L. S. Young and P. G. Murray, "Epstein-Barr virus and oncogenesis: from latent genes to tumours," *Oncogene*, vol. 22, no. 33, pp. 5108–5121, Aug. 2003, doi: 10.1038/sj.onc.1206556.

[94]. S. P. Lee, R. J. Tierney, W. A. Thomas, J. M. Brooks, and A. B. Rickinson, "Conserved CTL epitopes within EBV latent membrane protein 2: a potential target for CTL-based tumor therapy," *J Immunol*, vol. 158, no. 7, pp. 3325–3334, Apr. 1997.

[95]. S. P. Lee *et al.*, "HLA A2.1-restricted cytotoxic T cells recognizing a range of Epstein-Barr virus isolates through a defined epitope in latent membrane protein LMP2," *J Virol*, vol. 67, no. 12, pp. 7428–7435, Dec. 1993, doi: 10.1128/JVI.67.12.7428-7435.1993.

[96]. R. F. Jarrett *et al.*, "Detection of Epstein-Barr virus genomes in Hodgkin's disease: relation to age," *J Clin Pathol*, vol. 44, no. 10, pp. 844–848, Oct. 1991, doi: 10.1136/jcp.44.10.844.

[97]. K. W. Lo, K. F. To, and D. P. Huang, "Focus on nasopharyngeal carcinoma," *Cancer Cell*, vol. 5, no. 5, pp. 423–428, May 2004, doi: 10.1016/s1535-6108(04)00119-9.

[98]. Y.-P. Chen, A. T. C. Chan, Q.-T. Le, P. Blanchard, Y. Sun, and J. Ma, "Nasopharyngeal carcinoma," *Lancet*, vol. 394, no. 10192, pp. 64–80, Jul. 2019, doi: 10.1016/S0140-6736(19)30956-0.

[99]. L. S. Young, C. W. Dawson, K. W. Brown, and A. B. Rickinson, "Identification of a human epithelial cell surface protein sharing an epitope with the C3d/Epstein-Barr virus receptor molecule of B lymphocytes," *Int J Cancer*, vol. 43, no. 5, pp. 786–794, May 1989, doi: 10.1002/ijc.2910430508.

[100]. C. T. Lin, C. R. Lin, G. K. Tan, W. Chen, A. N. Dee, and W. Y. Chan, "The mechanism of Epstein-Barr virus infection in nasopharyngeal carcinoma cells," *Am J Pathol*, vol. 150, no. 5, pp. 1745–1756, May 1997.

[101]. H. L. Chen, M. M. Lung, J. S. Sham, D. T. Choy, B. E. Griffin, and M. H. Ng, "Transcription of BamHI-A region of the EBV genome in NPC tissues and B cells," *Virology*, vol. 191, no. 1, pp. 193–201, Nov. 1992, doi: 10.1016/0042-6822(92)90181-n.

[102]. D. P. Huang *et al.*, "A region of homozygous deletion on chromosome 9p21-22 in primary nasopharyngeal carcinoma," *Cancer Res*, vol. 54, no. 15, pp. 4003–4006, Aug. 1994.

[103]. K. W. Lo, D. P. Huang, and K. M. Lau, "p16 gene alterations in nasopharyngeal carcinoma," *Cancer Res*, vol. 55, no. 10, pp. 2039–2043, May 1995.

[104]. D. P. Huang *et al.*, "Loss of heterozygosity on the short arm of chromosome 3 in nasopharyngeal carcinoma," *Cancer Genet Cytogenet*, vol. 54, no. 1, pp. 91–99, Jul. 1991, doi: 10.1016/0165-4608(91)90035-s.

[105]. A. B. Hui *et al.*, "Loss of heterozygosity on the long arm of chromosome 11 in nasopharyngeal carcinoma," *Cancer Res*, vol. 56, no. 14, pp. 3225–3229, Jul. 1996.

[106]. G. Niedobitek, A. Agathanggelou, P. Barber, L. A. Smallman, E. L. Jones, and L. S. Young, "P53 overexpression and Epstein-Barr virus infection in undifferentiated and squamous cell nasopharyngeal carcinomas," *J Pathol*, vol. 170, no. 4, pp. 457–461, Aug. 1993, doi: 10.1002/path.1711700409.

[107]. Q. L. Lu, G. Elia, S. Lucas, and J. A. Thomas, "Bcl-2 proto-oncogene expression in Epstein-Barr-virus-associated nasopharyngeal carcinoma," *Int J Cancer*, vol. 53, no. 1, pp. 29–35, Jan. 1993, doi: 10.1002/ijc.2910530107.

[108]. M. T. Bejarano and M. G. Masucci, "Interleukin-10 abrogates the inhibition of Epstein-Barr virus-induced B-cell transformation by memory T-cell responses," *Blood*, vol. 92, no. 11, pp. 4256–4262, Dec. 1998.

[109]. Y. T. Huang *et al.*, "Profile of cytokine expression in nasopharyngeal carcinomas: a distinct expression of interleukin 1 in tumor and CD4+ T cells," *Cancer Res*, vol. 59, no. 7, pp. 1599–1605, Apr. 1999.

[110]. K. Sun *et al.*, "EBV-Positive Gastric Cancer: Current Knowledge and Future Perspectives," *Front. Oncol.*, vol. 10, p. 583463, Dec. 2020, doi: 10.3389/fonc.2020.583463.

[111]. B. Luo *et al.*, "Expression of Epstein-Barr virus genes in EBV-associated gastric carcinomas," *World J Gastroenterol*, vol. 11, no. 5, pp. 629–633, Feb. 2005, doi: 10.3748/wjg.v11.i5.629.

[112]. M. Sugiura *et al.*, "Transcriptional analysis of Epstein-Barr virus gene expression in EBV-positive gastric carcinoma: unique viral latency in the tumour cells," *Br J Cancer*, vol. 74, no. 4, pp. 625–631, Aug. 1996, doi: 10.1038/bjc.1996.412.

[113]. S. Imai *et al.*, "Gastric carcinoma: monoclonal epithelial malignant cells expressing Epstein-Barr virus latent infection protein," *Proc Natl Acad Sci U S A*, vol. 91, no. 19, pp. 9131–9135, Sep. 1994, doi: 10.1073/pnas.91.19.9131.

[114]. P. G. Murray *et al.*, "In situ detection of the Epstein-Barr virus-encoded nuclear antigen 1 in oral hairy leukoplakia and virus-associated carcinomas," *J Pathol*, vol. 178, no. 1, pp. 44–47, Jan. 1996, doi: 10.1002/(SICI)1096-9896(199601)178:1<44::AID-PATH471>3.0.CO;2-0.

[115]. T.-C. Cheng, S.-S. Hsieh, W.-L. Hsu, Y.-F. Chen, H.-H. Ho, and L.-F. Sheu, "Expression of Epstein-Barr nuclear antigen 1 in gastric carcinoma cells is associated with enhanced tumorigenicity and reduced cisplatin sensitivity," *Int J Oncol*, vol. 36, no. 1, pp. 151–160, Jan. 2010.

[116]. N. Sivachandran, C. W. Dawson, L. S. Young, F.-F. Liu, J. Middeldorp, and L. Frappier, "Contributions of the Epstein-Barr virus EBNA1 protein to gastric carcinoma," *J Virol*, vol. 86, no. 1, pp. 60–68, Jan. 2012, doi: 10.1128/JVI.05623-11.

[117]. H. Namba-Fukuyo *et al.*, "TET2 functions as a resistance factor against DNA methylation acquisition during Epstein-Barr virus infection," *Oncotarget*, vol. 7, no. 49, pp. 81512–81526, Dec. 2016, doi: 10.18632/oncotarget.13130.

[118]. D. H. Kim *et al.*, "Epstein-Barr virus BARF1-induced NFκB/miR-146a/SMAD4 alterations in stomach cancer cells," *Oncotarget*, vol. 7, no. 50, pp. 82213–82227, Dec. 2016, doi: 10.18632/oncotarget.10511.

[119]. M. S. Chang, H. S. Lee, E. J. Jung, C. W. Kim, B. L. Lee, and W. H. Kim, "Cell-cycle regulators, bcl-2 and NF-kappaB in Epstein-Barr virus-positive gastric

carcinomas," *Int J Oncol*, vol. 27, no. 5, pp. 1265–1272, Nov. 2005.
[120]. F. R. van Diemen *et al.*, "CRISPR/Cas9-Mediated Genome Editing of Herpesviruses Limits Productive and Latent Infections," *PLoS Pathog*, vol. 12, no. 6, p. e1005701, Jun. 2016, doi: 10.1371/journal.ppat.1005701.
[121]. R. Guo *et al.*, "DNA methylation enzymes and PRC1 restrict B-cell Epstein-Barr virus oncoprotein expression," *Nat Microbiol*, vol. 5, no. 8, pp. 1051–1063, Aug. 2020, doi: 10.1038/s41564-020-0724-y.
[122]. G. Gross, T. Waks, and Z. Eshhar, "Expression of immunoglobulin-T-cell receptor chimeric molecules as functional receptors with antibody-type specificity," *PNAS*, vol. 86, no. 24, pp. 10024–10028, Dec. 1989, doi: 10.1073/pnas.86.24.10024.
[123]. M. V. Maus, S. A. Grupp, D. L. Porter, and C. H. June, "Antibody-modified T cells: CARs take the front seat for hematologic malignancies," *Blood*, vol. 123, no. 17, pp. 2625–2635, Apr. 2014, doi: 10.1182/blood-2013-11-492231.
[124]. X. Tang *et al.*, "T cells expressing a LMP1-specific chimeric antigen receptor mediate antitumor effects against LMP1-positive nasopharyngeal carcinoma cells in vitro and in vivo," *J Biomed Res*, vol. 28, no. 6, pp. 468–475, Nov. 2014, doi: 10.7555/JBR.28.20140066.
[125]. H. Zimmermann and R. U. Trappe, "Therapeutic options in post-transplant lymphoproliferative disorders," *Ther Adv Hematol*, vol. 2, no. 6, pp. 393–407, Dec. 2011, doi: 10.1177/2040620711412417.
[126]. P. Feugier *et al.*, "Long-term results of the R-CHOP study in the treatment of elderly patients with diffuse large B-cell lymphoma: a study by the Groupe d'Etude des Lymphomes de l'Adulte," *J Clin Oncol*, vol. 23, no. 18, pp. 4117–4126, Jun. 2005, doi: 10.1200/JCO.2005.09.131.
[127]. P. Comoli *et al.*, "Preemptive therapy of EBV-related lymphoproliferative disease after pediatric haploidentical stem cell transplantation," *Am J Transplant*, vol. 7, no. 6, pp. 1648–1655, Jun. 2007, doi: 10.1111/j.1600-6143.2007.01823.x.
[128]. J. W. J. van Esser *et al.*, "Prevention of Epstein-Barr virus-lymphoproliferative disease by molecular monitoring and preemptive rituximab in high-risk patients after allogeneic stem cell transplantation," *Blood*, vol. 99, no. 12, pp. 4364–4369, Jun. 2002, doi: 10.1182/blood.v99.12.4364.
[129]. B. F. Israel, M. Gulley, S. Elmore, S. Ferrini, W. Feng, and S. C. Kenney, "Anti-CD70 antibodies: a potential treatment for EBV+ CD70-expressing lymphomas," *Mol Cancer Ther*, vol. 4, no. 12, pp. 2037–2044, Dec. 2005, doi: 10.1158/1535-7163.MCT-05-0253.
[130]. D. A. Thorley-Lawson and K. Geilinger, "Monoclonal antibodies against the major glycoprotein (gp350/220) of Epstein-Barr virus neutralize infectivity," *Proc Natl Acad Sci U S A*, vol. 77, no. 9, pp. 5307–5311, Sep. 1980, doi: 10.1073/pnas.77.9.5307.
[131]. J. I. Cohen, "Epstein-barr virus vaccines," *Clin Transl Immunology*, vol. 4, no. 1, p. e32, Jan. 2015, doi: 10.1038/cti.2014.27.
[132]. J. Snijder *et al.*, "An Antibody Targeting the Fusion Machinery Neutralizes Dual-Tropic Infection and Defines a Site of Vulnerability on Epstein-Barr Virus," *Immunity*, vol. 48, no. 4, pp. 799-811.e9, Apr. 2018, doi: 10.1016/j.immuni.2018.03.026.
[133]. D. Watterson *et al.*, "A generic screening platform for inhibitors of virus induced cell fusion using cellular electrical impedance," *Sci Rep*, vol. 6, p. 22791, Mar. 2016, doi: 10.1038/srep22791.