Trends in Molecular Medicine

CellPress OPEN ACCESS

Review

The Kaposi's sarcoma progenitor enigma: KSHV-induced MEndT–EndMT axis

Julian Naipauer [1,2,3,4,*] and Enrique A. Mesri[2,3,4,†]

Endothelial-to-mesenchymal transition has been described in tumors as a source of mesenchymal stroma, while the reverse process has been proposed in tumor vasculogenesis and angiogenesis. A human oncogenic virus, Kaposi's sarcoma herpes virus (KSHV), can regulate both processes in order to transit through this transition 'boulevard' when infecting KS oncogenic progenitor cells. Endothelial or mesenchymal circulating progenitor cells can serve as KS oncogenic progenitors recruited by inflammatory cytokines because KSHV can reprogram one into the other through endothelial-to-mesenchymal and mesenchymal-to-endothelial transitions. Through these novel insights, the identity of the potential oncogenic progenitor of KS is revealed while gaining knowledge of the biology of the mesenchymal-endothelial differentiation axis and pointing to this axis as a therapeutic target in KS.

Highlights

Endothelial-to-mesenchymal (EndMT) and mesenchymal-to-endothelial (MEndT) transitions have been described in tumors as a source of mesenchymal stroma and tumor vasculogenesis/angiogenesis, respectively.

Kaposi's sarcoma herpes virus (KSHV) can reprogram endothelial or mesenchymal cells one into the other after infection through endothelial-to-mesenchymal or mesenchymal-to-endothelial transitions.

KS remains potentially life-threatening for patients with advanced or antiretroviral-therapy-resistant KS.

The identification of the KS progenitor primary cell type and the environmental conditions leading to sarcomagenesis is still under debate. These would allow the development of more accurate and physiologically relevant KS animal models to test new-targeted therapies for KS.

KSHV induces the mesenchymal–endothelial differentiation axis in KS progenitor cells, indicating the importance of this mechanism in viral oncogenesis.

The KS progenitor enigma

Transformation of epithelial cells into mesenchymal cells (epithelial–mesenchymal transition, EMT) regulates gastrulation, neural crest, somite dissociation, craniofacial development, wound healing, organ fibrosis, and tumor metastasis [1]. The EMT program induces carcinoma cells to lose their adhesion, acquire mesenchymal properties, and migrate away from the primary tumor [2]. Endothelial cells can undergo a similar process termed **endothelial to mesenchymal transition (EndMT)** (see Glossary), that is critical for endocardial cushion formation, cardiac and kidney fibrosis, atherosclerosis, tissue remodeling and, in the tumor microenvironment, generates carcinoma-associated fibroblasts (CAFs) that may be essential for cancer progression [3–5]. **Mesenchymal to endothelial transition (MEndoT**) is the reverse form of EndMT and is involved in **vasculogenesis** and **angiogenesis** scenarios [6,7]. Recently, this process has also been linked to a source of endothelial cells in the tumor microenvironment [8,9]. Cumulative evidence shows that a human oncovirus, **Kaposi's sarcoma herpesvirus (KSHV)**, can regulate both pathways for transiting through this EndMT - MEndoT 'boulevard' when infecting the potential oncogenic progenitor cell of **Kaposi's sarcoma (KS)** depending on whether it belongs to the endothelial or to the mesenchymal lineage. Evidence that this virus can induce both EndMT and MEndoT supports the importance of these transitions as a mechanism of **viral oncogenesis**. Moreover, the cell type heterogeneity in the KS lesions and the fact that KSHV can infect a variety of cell types, including endothelial and mesenchymal cells, makes the identification of the KS cell origin more challenging. Identifying the KS progenitor primary cell types and the environmental conditions leading to Kaposi's **sarcomagenesis** would allow the development of more accurate and physiological relevant KS cell and animal models to test new-targeted therapies for KS and to model the different pathogenic outcomes of the disease.

In the present Review, we focus on the enigma of the KS progenitor, and efforts to identify this precursor through *in vitro* infection, tumorigenesis studies, and clinical findings, as a platform to

[1]Instituto de Fisiología, Biología Molecular y Neurociencias (IFIBYNE), CONICET-Universidad de Buenos Aires, Buenos Aires, Argentina
[2]Tumor Biology Program, Sylvester Comprehensive Cancer Center, University of Miami Miller School of Medicine, Miami, FL, USA
[3]University of Miami- Center for AIDS Research (UM-CFAR)/Sylvester Comprehensive Cancer Center (CCC) Argentina Consortium for Research and Training in Virally Induced AIDS-Malignancies, University of Miami Miller School of Medicine, Miami, FL, USA
[4]Miami Center for AIDS Research, Department of Microbiology and Immunology, University of Miami Miller School of Medicine, Miami, FL, USA

gain insight into the molecular and cellular mechanisms operating within the mesenchymal to endothelial transition reversible axis.

EndMT reversible axis

The EndMT process is characterized by profound morphological, functional, and molecular changes in the endothelial cell phenotype. During EndMT, endothelial markers are downregulated, while mesenchymal markers are *de novo* expressed by endothelial cells. These markers include FSP-1, α-smooth muscle actin (α-SMA), fibronectin, collagenase I, vimentin, and N-cadherin. After these changes have occurred, the endothelial cells lose their adhesion and change their morphology forming elongated, spindle-shaped cells that are highly invasive and migratory [3,10]. Transforming growth factor (TGF)β, bone morphogenetic protein (BMP), and Notch pathways are essential for stimulating the EndMT program, mediated by Smad-dependent and Smad-independent signaling pathways [11,12]. Transcription factors such as Snail, Slug, ZEB-1, SIP-1, Twist, and LEF-1 are linked to the loss of cell–cell adhesion by endothelial cells undergoing EndMT [12]. Metabolic and environmental conditions in tumors, such as oxidative stress and hypoxia, are conducive to EndMT. As tumors grow and become more hypoxic, this condition would increase CAFs that are generated by EndMT within the tumor microenvironment [10,13], and play an important role in supporting tumor growth. The conversion of endothelial cells into mesenchymal cells via EndMT is not an irreversible shift between two alternative cellular differentiation phenotypes. Recent investigations showed reversibility in this transitional process, implying that cells could remain in intermediary stages of differentiation and may frequently undergo a partial EndMT program [12]. Moreover, mesenchymal stem cells (MSCs) have been shown to revert to EndMT by fully undergoing MEndT. This inverse process is essential for **tumor angiogenesis** and promotes a proangiogenic niche through IGF-1 and p38 signaling pathways [9,14] (Figure 1). Moreover, a study of clinical KS specimens has provided evidence for MEndT in KS, adding to the evidence fully discussed in the following section that KS may also originate from MSCs and cannot be considered only an endothelial lineage originated tumor [15].

Mechanisms of oncogenesis driven by KSHV gene expression in KS

In 1872, the Hungarian physician and dermatologist Moritz Kaposi described several cases of a multifocal-pigmented sarcoma of the skin in elderly European men. In 1994, KSHV or human herpesvirus-8 (HHV-8) was discovered and found to be the etiological agent of KS [16]. Since then, this virus remains the subject of intense research aimed at understanding its oncogenic mechanisms since KS remains a clinical challenge, particularly in AIDS and low-resource settings. Furthermore, KSHV infection induces the reprogramming of gene expression and morphological changes in infected cells [17,18]. The first recognizable presentation of KS is the patchy lesions that display inflammatory cells and angiogenesis with spindle-shaped cells that later dominate the lesions. As spindle cells proliferate, the lesions progress to predominantly spindle cells with inflammatory cells and intense angiogenesis as well [19]. Explanted, KS spindle cells that lose KSHV differ from most other tumor cells as they depend on external cytokines and growth factors to grow *in vitro* and do not induce tumors in nude mice, unlike truly transformed cells [20–22]. However, in the presence of inflammatory cytokines, cells isolated from KS lesions can induce angiogenic lesions in immunodeficient mice [20,23], further indicating the importance of inflammation in developing KS. KS remains potentially life-threatening for patients with advanced or antiretroviral therapy (ART)-resistant KS, where systemic therapy is indicated, and three FDA-approved agents that include liposomal anthracyclines are available [24–26]. Despite the effectiveness of these agents, some patients develop ART-resistant KS requiring additional therapy [27].

KSHV infection is essential for KS development, yet it is not sufficient as KS development occurs in people with other risk factors, including aging [24]. Like other members of the Herpesviridae

†Deceased, August 27, 2022

*Correspondence:
juliannaipauer@gmail.com (J. Naipauer).

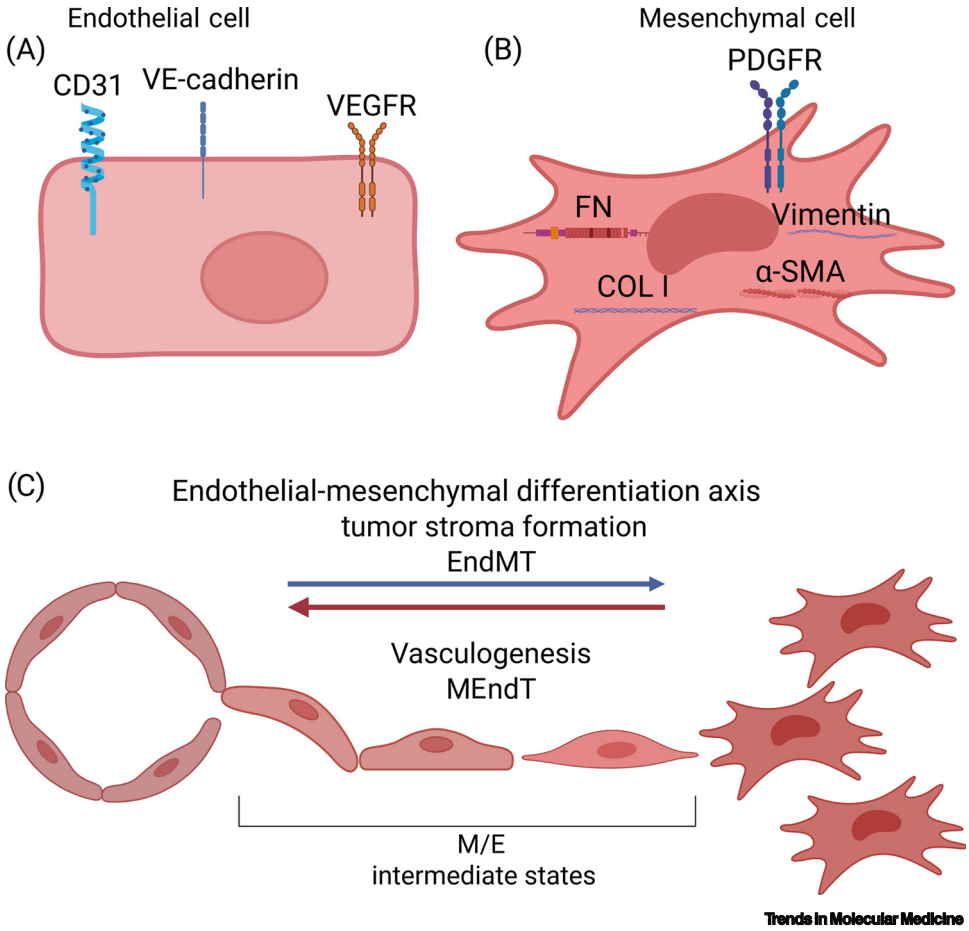

Figure 1. Endothelial–mesenchymal differentiation axis. (A) Scheme of an endothelial cell showing specific markers: CD31 (PECAM-1), VEGFR, and VE-cadherin. (B) Scheme of a mesenchymal cell showing specific markers: PDGFR, FN (fibronectin), COL I (collagen type I), α-SMA (α-smooth muscle actin), and vimentin. (C) Endothelial–mesenchymal differentiation axis proposed mechanism for transitions: Endothelial-to-mesenchymal transition (EndMT) through tumor stroma formation and mesenchymal-to-endothelial transition (MEndT) through vasculogenesis. The mesenchymal/endothelial intermediate states (M/E) are also depicted. Created with BioRender.com.

family, HHV-8/KSHV displays a biphasic life cycle with a latent and a lytic phase. Latency represents the default mode of persistence in the host cells with the expression of a few viral genes and viral miRNAs involved in the maintenance of the viral episome and the KSHV-induced tumorigenesis [28]. During the lytic phase, KSHV expresses many viral genes involved in viral replication and particle production that eventually end in cellular lysis [29]. Both latent and lytic phases contribute to KS pathogenesis [29]. However, it is unclear how these two viral life phases coexist and interact in *de-novo*-infected progenitor cells to induce the entire KSHV transformation program. Despite the low level of KSHV lytic reactivation in KS tumors, it appears that this phase is crucial for the virus-induced tumorigenesis, demonstrated by the high viral loads in plasma associated with the worst KS prognosis and by the efficacy of lytic replication inhibitors (i.e., ganciclovir and foscarnet) on preventive KS recurrence [30,31]. Furthermore, lytic replication would be needed for virus production to maintain the population of infected cells in the affected tissue.

Overexpression of KSHV oncogenes can induce KS-like tumor formation in mice, pointing to these genes as significant contributors to KSHV sarcomagenesis [24,29,32–36]. A paracrine

Glossary

Angiogenesis: growth of blood vessels from the existing vasculature. It occurs throughout life in health and disease, beginning in utero and continuing through old age.

Endothelial to mesenchymal transition (EndMT): a biological process in which endothelial cells adopt a mesenchymal phenotype displaying typical mesenchymal cell morphology and functions. Recent findings have shown that EndMT is related to resistance to cancer therapy, such as chemotherapy, antiangiogenic, and radiation therapy.

Kaposi's sarcoma (KS): cancer that usually appears as tumors on the skin or mucosal surfaces such as inside the mouth, but these tumors can also develop in other parts of the body, such as in the lymph nodes (bean-sized collections of immune cells throughout the body), the lungs, or digestive tract.

Kaposi's sarcoma herpesvirus (KSHV): also known as human herpesvirus 8; the etiological agent of KS. KSHV also causes primary effusion lymphoma and multicentric Castleman's disease.

Mesenchymal to endothelial transition (MEndT): a biological process by which mesenchymal cells transform into endothelial cells. This process was found in cardiac fibroblasts that acquired an endothelial cell phenotype, promoted angiogenesis, improved cardiac repair, and inhibited cardiac fibrosis. Moreover, MEndT was also reported in juvenile angiofibromas and KS.

Receptor tyrosine kinase (RTK): a subclass of TKs that are involved in mediating cell-to-cell communication and controlling a wide range of complex biological functions, including cell growth, motility, differentiation, and metabolism. Dysregulation of RTK signaling leads to many human diseases, especially cancer.

Sarcomagenesis: sarcomas comprise a heterogeneous group of malignancies derived from mesenchymal cells, which under normal circumstances lead to the development of connective tissues such as bone, muscle, fat, and cartilage. During the past decade, insight has been gained regarding the aberrancies that occur during normal development that result in mesenchymal cells transforming into sarcomas.

oncogenesis hypothesis has been proposed to explain the interaction between latent and lytic gene expression and its regulation of infected and uninfected cells, where the lytic KSHV-induced secretion of proinflammatory and angiogenic factors would modulate the behavior of latently infected cells, and potentially uninfected cells, in the tumor microenvironment [24,37–39]. Supporting this hypothesis, Montaner *et al.* showed that cells expressing KSHV lytic genes enable tumorigenicity of latently infected cells via paracrine mechanisms [40]. Another hypothesis relies on the existence, in some cellular contexts, of a dysregulated viral transcriptional program that is neither latent nor lytic, called the abortive lytic phase [41]. This phase is characterized by sporadic expression of lytic genes without efficient production of infectious particles. This pattern of KSHV gene expression was observed in endothelial lineage cells transduced with a KSHV bacterial artificial chromosome (KSHVBac36) to generate a cell line (mECK36) that is able to form KS-like tumors in mice [32]. This was also observed upon KSHV infection of murine bone-marrow-derived (BM)-MSCs growing in a proangiogenic environment [35]. A possible mechanism described to explain this abortive phase in BM-MSCs is the epigenetic regulation of a derepressed KSHV and host epigenome leading, on the one hand, to upregulation of oncogenic genes that include viral lytic genes and host genes, including platelet-derived growth factor (PDGF), vascular endothelial growth factor (VEGF), Toll-like receptor signaling, and p53 downregulation; and on the other hand, the most epigenetically repressed pathways were linked to innate immunity. This epigenetic pattern could favor the ability of these cells to continue growing while repressing innate immune genes that would curtail growth [35]. A similar viral transcriptional program has also been described in human lymphatic endothelial cells infected with KSHV [39], suggesting that this mechanism also occurs in primary human cells. Another crucial aspect of the interplay between latent and lytic KSHV gene expression in KSHV-induced tumorigenesis is further reinforced by the fact that many autocrine and paracrine interactions operate within KS tumors. These KSHV-induced ligand-dependent interactions include activating the VEGF–VEGF receptor (VEGFR)2 axis [34] and activating PDGF receptor α (PDGFRA) signaling through the upregulation of PDGFs by KSHV lytic genes that can drive the KSHV tumorigenesis [33]. This activation could act in a paracrine manner from either lytically or abortive lytic KSHV-infected cells expressing VEGF and PDGF upregulated by viral oncogenes, such as vGPCR and K1, to induce KSHV tumorigenesis [29,37]. Both are plausible possibilities, viral oncogenes expressed from lytic or abortive lytic cells, and would explain the reprogramming of the KS spindle cell progenitors to help achieve the hallmarks of cancer needed to develop the KSHV-induced sarcomagenesis [42].

Tumor angiogenesis: proliferation of a network of blood vessels that penetrates cancerous growths, supplying nutrients and oxygen and removing waste products. Tumor angiogenesis starts with cancerous tumor cells releasing molecules that send signals to surrounding normal host tissue.
Vasculogenesis: creation of new blood vessels; involves differentiation of angioblasts and is responsible for the basic formation of a vascular network. Vasculogenesis mainly occurs only during embryonic development.
Viral oncogenesis: the feature of tumor viruses that induces benign or malignant proliferation of infected cells. Viruses can infect cells and manipulate the cell's machinery to create more viruses. During this viral replication process, certain viruses affect the host cell's genes so that they may cause it to become much more cancerous.

The case for an endothelial cell lineage as KS spindle cell origin

The ontogeny of KS is further complicated as KS presents as multifocal lesions. Furthermore, the multiclonal malignant nature and diversity of markers described by several groups make the spindle cells even more elusive. Spindle cells from KS lesions are poorly differentiated and express different markers corresponding to different lineages such as endothelial, smooth muscle, dendritic cell, macrophage, and mesenchymal markers (Table 1) [29,33,43–47]. Based on the conspicuous

Table 1. Markers from different cell lineages expressed by KS spindle cells

Cell lineage	Marker	Refs
Endothelial	CD31/PECAM1, CD34, factor VIII, D2-40, VEGFR3, VEGFR2, LYVE-1, CD105/endoglin and PDPN/podoplanin	[29,43–45,96]
Macrophages	CD68	[46]
Dendritic	Factor XIII	[46]
Smooth muscle	SMA	[47,78,96]
Mesenchymal	PDGFR, nestin, vimentin and CD29	[33,78,96,98]

expression of microvascular endothelial markers in the KS lesions and the spindling of endothelial cells upon infection or inflammatory cytokine exposure [20,48–50], the first emerging theory to explain the occurrence of KS spindle cells was its endothelial cell (EC) lineage origin. Various models of primary EC infection by KSHV showed the importance of these cells in KSHV oncogenesis and tumorigenesis [28,51]. Early reports showed a long-term persistent infection of primary ECs by KSHV, although they could not show complete transformation phenotypes [20,48–50,52–54]. To overcome this issue, several models have been developed to immortalize endothelial cells before KSHV infection and have shown to be valuable tools for studying KSHV oncogenesis. Immortalized dermal microvascular endothelial cells with the E6 and E7 genes from human papillomavirus type 16 before KSHV infection showed latent infection with only a small fraction of cells expressing lytic genes, production of infectious viral progeny, and transformed characteristics, including loss of contact inhibition and acquisition of anchorage-independent growth [55]. Human umbilical vein endothelial cells (HUVECs) conditionally immortalized by lentiviral transfer of doxycycline controlled SV40 large T antigen and hTERT expression cassettes (HuARLT cells), infected with KSHV become spindle-like, lose endothelial properties, and undergo transcriptional changes corresponding to EndMT [56–58]. Dermal microvascular endothelial cells immortalized with a retrovirus expressing the telomerase reverse transcriptase subunit (hTERT), TIME cells, showed KSHV latent infection. Such cultures could be induced to the lytic phase, generating infectious progeny that can be serially propagated *in vitro* [59]. hTERT-immortalized HUVEC line (TIVE) infected with KSHV can transform and form KS-like tumors in immunocompetent mice [60,61]. Using these models, several host cellular and viral genes were identified to have a role in the transformation program induced by KSHV [62–71]. These results point to microvascular ECs found in the lesions as important contributors to the oncogenic program triggered by KSHV.

KSHV infection of primary blood vascular endothelial cells (BECs) or primary lymphatic endothelial cells (LECs) results in different viral expression programs; while the virus is latent in BECs, it displays a spontaneous lytic replication program in LECs [39,72]. KSHV infection of LECs drives their dedifferentiation toward the BEC phenotype, while KSHV infection of BECs drives their differentiation toward the LEC phenotype [43,44,73,74], demonstrating the remarkable reprogramming capacity of KSHV infection. Recently, two papers have shown that while KSHV-infected BECs display a strong tendency to viral loss as they proliferate, KSHV-infected LECs are inefficient in establishing latency and initiating the productive lytic replication program with the release of infectious viral particles and cell death [75,76]. However, the lymphatic cell environment; directed by the expression of SOX18, which promotes an increase in the number of latent genomes; and PROX1, which directly stimulates the expression of RTA/ORF50, might provide infectious viral particles and lytic chemokines for *de novo* infection of KS progenitor cells [75,76]. Moreover, KSHV-infected primary ECs would contribute to KS development through a first stage involving EndMT to support the progression of the disease through the spread of the infected cells into the surrounding connective tissue [77]. Cheng *et al.* showed, using a 3D cell model for KSHV infection of LECs, that KSHV induces transcriptional reprogramming of lymphatic endothelial cells to invasive mesenchymal cells via EndMT, triggering KSHV-induced Notch-dependent signaling, leading to PDGFR expression and MT1–MMP-dependent invasiveness, pointing to this mechanism as important for KSHV-induced sarcomagenesis [78,79]. Moreover, this 3D system reproduces patterns of KSHV gene expression and the presence of mesenchymal KSHV-infected cells found in KS lesions (Figure 2).

The case for circulating precursor cells as KS progenitors

Circulating KS progenitors such as endothelial progenitor cells (EPCs) and MSCs can be potential KS oncogenic progenitors [18]. A first report on the possible origin of the KS progenitor from circulating cells was published by Browning *et al.* in 2004. They showed that cells displaying similar

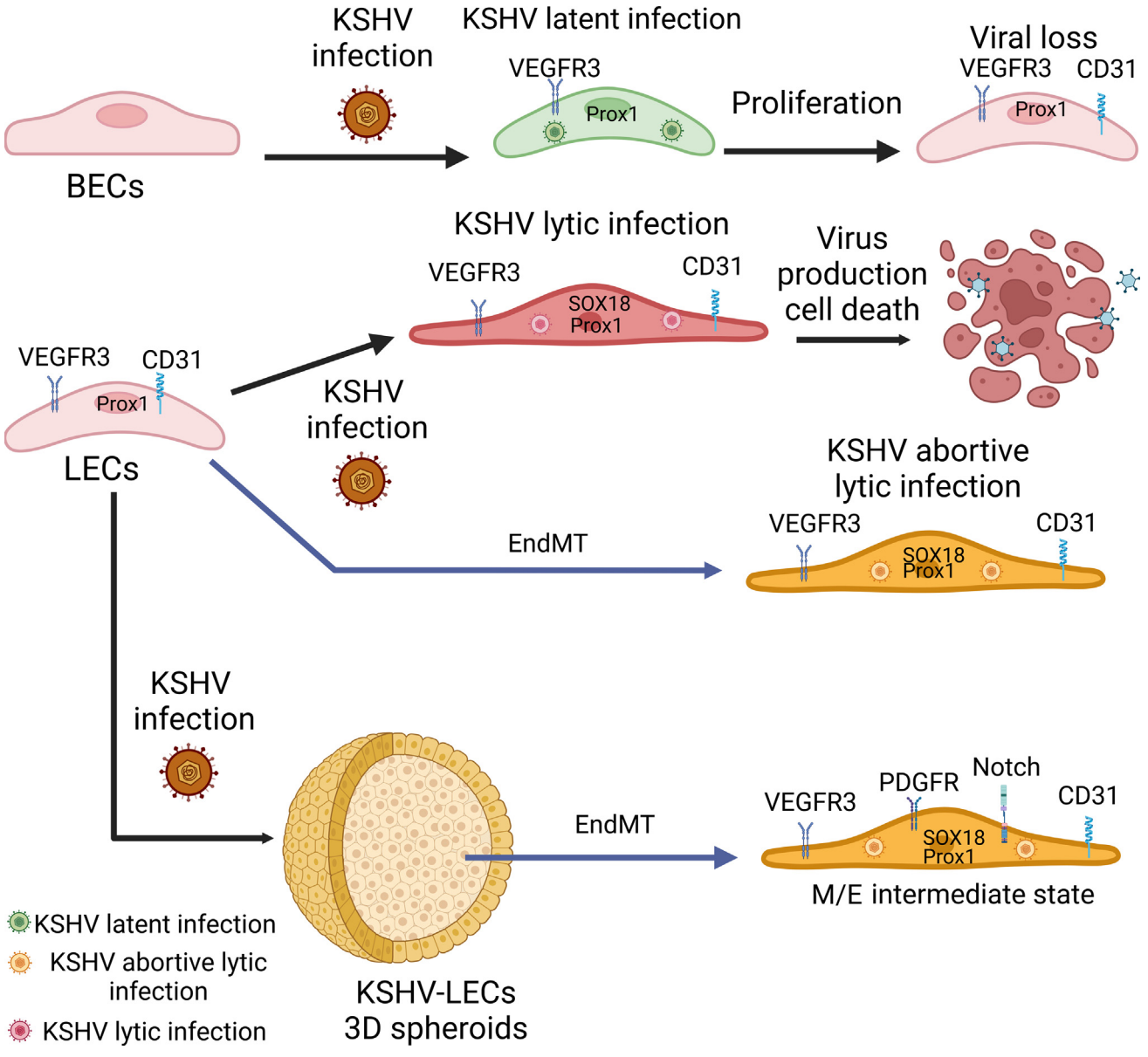

Figure 2. Endothelial cell origin for Kaposi's sarcoma (KS) spindle cells. KS herpesvirus (KSHV) infection of primary blood vascular endothelial cells (BECs) or primary lymphatic endothelial cells (LECs) results in different viral expression programs with different outcomes; while the virus is latent in BECs with viral loss, it displays a spontaneously lytic replication program in LECs. When growing in 3D spheroids KSHV reprograms LECs to invasive mesenchymal cells via endothelial-to-mesenchymal transition (EndMT), generating mesenchymal/endothelial (M/E) intermediate state cells with KSHV abortive lytic infection. CD31 is an endothelial marker. VEGFR3, SOX18, and PROX1 are markers of lymphatic endothelium. PDGFR and Notch are mesenchymal stem cell markers. Created with BioRender.com.

markers to those of spindle cells from KS lesions of AIDS patients could be obtained from the peripheral blood by exposure to inflammatory cytokines from activated T cell conditioned medium (CM). This CM contains the same inflammatory cytokines increased in HIV-1-infected individuals. These cells showed phenotypic and functional similarities with AIDS–KS spindle cells, including expression of vWF, vimentin, SMA, and the ability to induce angiogenesis *in vivo* [23]. Inflammatory cytokines can recruit and differentiate potential circulating KS progenitors inducing spindle

cell differentiation and proliferation, which correlate with the propensity of KS lesions to localize to scar tissue or sites of inflammation growing in a cytokine-rich microenvironment [80]. EPCs are increased in the peripheral blood of patients with KS [81]; circulating EPCs isolated from KS patients were positive for KSHV, retained the virus, and were able to sustain lytic replication [82]. Moreover, KS spindle cells in renal transplant recipients are of donor origin [83], thus suggesting that infected circulating cells can serve as KS progenitors.

KS models made from circulating mouse endothelial lineage derived from bone marrow [32,84] and KSHV-infected primary rat embryonic metanephric circulating mesenchymal precursor cells [85] showed KS-like tumor formation in nude mice. Still, neither of these studies fully identified lineage-specific markers for the circulating primary progenitor cell type to be transformed by KSHV infection. However, these models of KSHV-induced tumorigenesis suggest that these populations contain specific cell types in which KSHV infection is oncogenic, supporting the theory of circulating KS progenitor cells. Furthermore, circulating EPCs can undergo EndMT [86] and can be important for KSHV-induced oncogenesis through this transition axis.

The case for MSCs as circulating KS progenitors

Sarcomas are tumors of mesenchymal cell origin. KS might not be the exception, as most KS lesions express the MSC-defining marker PDGFRA [33]. MSCs are essential components of BM precursor cells, multipotent undifferentiated stem cells that can differentiate into various mesodermal cells, such as osteoblasts, chondrocytes, and adipocytes. Still, they can also differentiate into nonmesodermal cells, such as ECs [87]. The transformation of MSCs by different mechanisms gives rise to the formation of sarcomas *in vivo*, placing MSCs as the cells of origin for sarcomas [88].

Two studies have shown the susceptibility of fetus-derived human MSCs to KSHV infection *in vitro* [89] and the enhanced angiogenic potential of MSCs after KSHV infection [90]. Lee *et al.* showed that KSHV could efficiently infect human MSCs of diverse origin. KSHV-infected human MSCs acquire KS-like cell surface markers and angiogenic, invasive, and transforming phenotypes [91]. It was found that KSHV infection of BM-MSCs in MSC culture conditions favors viral production but hinders infected cell proliferation. In contrast, proangiogenic KS-like culture conditions are more permissive to the proliferation of productively infected human MSC cultures [35]. KSHV infection of oral MSCs induced MEndT and enhanced the expression of a large number of chemokines (CCL5, CCL8, CXCL10, etc.), inflammatory cytokines (IL1, IL6, TNFSF10, etc.), and angiogenic factors (bFGF, VEGF, PGF, ANGPTL2, etc.); some of which are essential for KSHV-induced tumorigenesis [92]. Compared with KS lesions with a KS gene signature, KSHV-infected oral MSC clusters close to KS lesions more than KSHV-infected ECs, including HMVECs, LECs, and BECs [92], indicating that KSHV-infected MSCs would recapitulate better the gene expression profile found in KS lesions. Wang *et al.* showed that KSHV infection of human MSCs significantly promoted MSCs migration and homing to wound sites [93]. Chen *et al.* found an association between human osteosarcoma and KSHV [94], reinforcing the idea of KSHV as a sarcomagenic virus. KSHV activates *PROX1* gene expression and initiates MEndT in human MSCs, rendering MSC tumorigenic features such as angiogenesis, invasion, and migration [95]. Chen *et al.* showed that KSHV infection of MSCs initiates an incomplete MEndT process and generates hybrid mesenchymal/endothelial (M/E) state cells and that KS lesions contained a large number of tumor cells with a M/E state that may result from an incomplete MEndT or EndMT [96]. Together, these data reinforce the hypothesis that KS may derive from circulating KSHV-infected MSCs migrating into an inflammatory and angiogenic site, inducing MEndT and enhancing the transformation capacity of KSHV. At the same time, viral production by infected MSCs can also serve as a source of virus for infection of new

migrating uninfected progenitor cells. Moreover, the transformation of mouse PDGFRA-positive BM-MSCs infected with KSHV happens only in angiogenic KS-like conditions [35], reinforcing the idea that these specific cell types are the putative primary circulating KS progenitor cell in a proangiogenic environment (Figure 3).

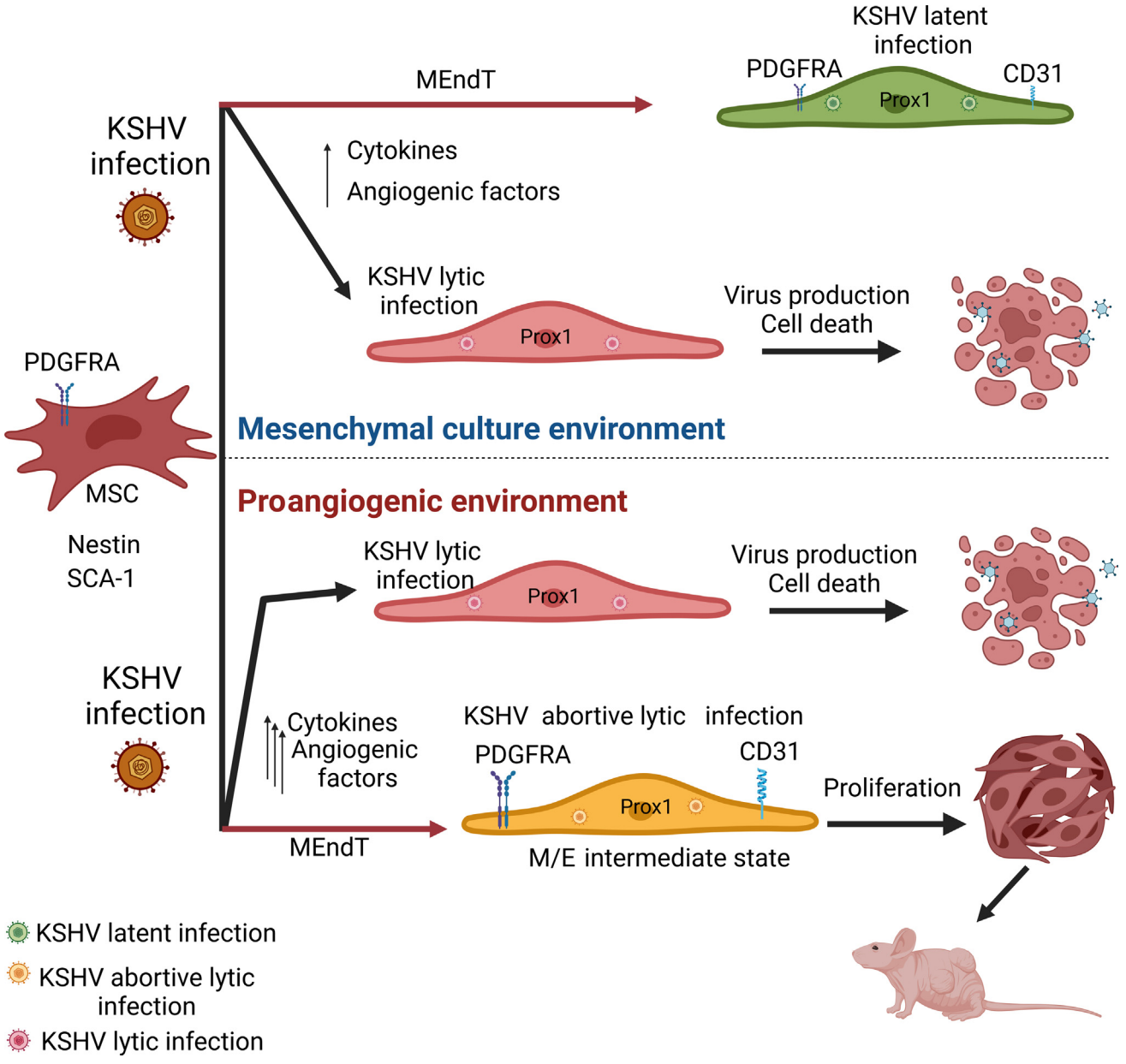

Figure 3. Mesenchymal cell origin for Kaposi's sarcoma (KS) spindle cells. KS herpesvirus (KSHV)-infected human mesenchymal stem cells (MSCs) can acquire KS-like cell surface markers and angiogenic, invasive, and transforming phenotypes. (Top) While mesenchymal culture conditions induce mesenchymal-to-endothelial transition (MEndT) with latent infection, this results in proliferation arrest or favors viral production with cell death of infected cells. (Bottom) By contrast KS-like proangiogenic environments are more permissive for KSHV abortive lytic infection enabling the proliferation of productively infected human MSC cultures and inducing MEndT, which renders mesenchymal/endothelial (M/E) intermediate states and enhanced transformation capacity of KSHV. Nestin, SCA-1, and PDFRA are MSC markers. PROX1 is a lymphatic endothelium marker. CD31 is an endothelial marker. Created with BioRender.com.

PDGFRA as KS MSC progenitor defining marker and oncogenic driver in KS

The KS spindle-cell progenitor identity, defining markers, and specific host conditions of KSHV-driven oncogenesis upon *de novo* infection are still under debate (Table 2). Early reports suggest that KS is an angiogenic and inflammatory cytokine-mediated driven disease, at least in the early stages, and that angiogenic factors play a role in lesion development [21,22,97]. BM-derived PDGFRA/SCA-1 expressing MSCs were identified as potential KS spindle-cell progenitors. It was found that proangiogenic environmental conditions characteristic of KS inflammation and wound healing are critical for KSHV sarcomagenesis [35]. These growth conditions allow

Table 2. KS spindle progenitor cell type evidence

Progenitor cell	Evidence	Refs
Endothelial cell lineage	•Long-term persistent infection of primary endothelial cells by KSHV	[49,50,52,53]
	•Reprograming of BECs and LECs after KSHV infection	[43,44,73]
	•BECs display a strong tendency for viral loss as they proliferate	[75,76]
	•LECs lytically infected might provide infectious viral particles and lytic chemokines for de novo infection of KS progenitor cells	[75,76]
	•Primary endothelial cells could contribute to KS development through a first stage involving EndMT	[77,78]
Mesenchymal stem cells	•Association of human osteosarcoma with KSHV	[94]
	•Evidence for MEndT in KS	[15]
	•Enhanced angiogenic potential of MSCs after KSHV infection	[89,90]
	•KSHV-infected human MSCs acquire KS-like cell surface markers and angiogenic, invasive, and transforming phenotypes	[91]
	•KSHV-infected human MSCs in MSC culture conditions favor viral production with oncogene-induced senescense	[35]
	•KSHV-infected human MSCs in proangiogenic KS-like culture conditions are more permissive for enabling proliferation	[35]
	•KSHV infection of human MSC-induced MEndT, enhances expression of chemokines, inflammatory cytokines, and angiogenic factors	[92]
	•KSHV-infected oral MSCs cluster closer to KS lesions than KSHV-infected endothelial cells HMVECs, LECs, and BECs	[92]
	•KSHV infection of human MSCs significantly promoted MSCs migration and settlement in the wound sites	[93]
	•KSHV activates PROX1 gene expression and initiates MEndT in human MSCs	[95]
	•KSHV infection of MSCs initiates an incomplete MEndT process and generates hybrid M/E state cells	[96]
	•Transformation capacity of mouse PDGFRA-positive BM-MSCs infected with KSHV only in KS-like conditions	[35]
Circulating progenitors	•KS-like spindle cells can be cultured from the peripheral blood of KS patients using inflammatory cytokines from activated lymphocytes	[23]
	•Circulating progenitors are increased in the peripheral blood of patients with KS	[81]
	•Circulating progenitors isolated from KS patients were found positive for KSHV, retained the virus, and were able to sustain lytic replication	[82]
	•KS spindle cells in renal-transplant recipients have been found to be from donor origin	[83]
	•KS models made from mouse endothelial lineage derived from bone marrow showed KS-like tumor formation in nude mice	[32,84]
	•KS model made from KSHV-infected primary rat embryonic metanephric mesenchymal precursor cells showed KS-like tumor formation in nude mice	[85]

Clinician's corner

KSHV or HHV-8 is the etiological agent of KS.

KS occurs in the context of aging in specific ethnic populations due to chronic inflammation and transplant-associated immune suppression and is the most common cancer in HIV-AIDS patients.

Systemic therapy is indicated, and three FDA-approved agents, including liposomal anthracyclines, are available. Despite the effectiveness of these agents, most patients progress and require additional therapy.

Viruses tend to coopt oncogenic pathways as persistence and replication strategies, which tend to be dysregulated in nonviral cancers of the exact cell origin.

In a Phase 2 trial, >50% of AIDS-KS patients benefited from imatinib mesylate (Gleevec), an FDA-approved PDGFR inhibitor.

Sunitinib, a drug approved for renal cell carcinoma and tested in Phase 1 clinical trials for tumors in the context of HIV/AIDS, showed in mouse KS tumors a more potent antitumor effect than imatinib. This correlated with complete blockage of PDGFR signaling and more anti-angiogenic activity since it is able to target VEGF receptors, indicating that complete inhibition of RTKs signaling has therapeutic benefits in AIDS-KS.

The importance of the mesenchymal–endothelial differentiation axis in the process of cancer and viral oncogenesis opens the door for testing strategies to inhibit this differentiation axis as a new therapeutic approach for KS.

KSHV-infected, PDGFRA-positive MSCs to overcome KSHV-driven oncogene-induced senescence and cell cycle arrest via a PDGFRA-signaling mechanism [35]. PDGF ligands and receptors are expressed in the AIDS-KS lesions [98,99] and can induce KS spindle cell proliferation [100,101]. Activated PDGFRA is the most predominantly activated oncogenic receptor tyrosine kinase (RTK) in mouse KS-like tumors, and it is consistently overexpressed and phosphorylated in KS lesions [33]. Together with imatinib studies in animals and clinical trials, these data show that KSHV-induced ligand-mediated activation of PDGFRA signaling appears to drive KS tumorigenesis [33]. These data indicate that PDGFRA is likely an oncogenic driver, target, and marker of AIDS-KS. Since viruses tend to coopt oncogenic pathways as persistence and replication strategies, which tend to be dysregulated in nonviral cancers of the exact cell origin, it follows that PDGFRA-expressing cells should be a preferred oncogenic target of KSHV [35,42], pointing to uninfected PDGFRA expressing circulating MSC as KS progenitors. In KS samples, Chen *et al.* recently showed that LANA-positive, spindle-shaped cells expressed MSC markers (nestin, PDGFRA, or α-SMA) and endothelial markers (PDPN, CD31, or VEGFR2) simultaneously and that the proportions of $LANA^+$ $PDGFRA^+$ $PDPN^+$ cells increased with KS progression [96]. These findings are consistent with the mesenchymal origin of KS, which is similar to nonviral sarcomas driven mainly by PDGFRA dysregulation, caused by mutation or ligand stimulation [88,102,103]. PDGFRA expression in KS lesions reinforces the histopathological classification of KS as a true sarcoma. In a Phase 2 trial, >50% of patients benefited from imatinib mesylate (Gleevec), an FDA-approved PDGFR inhibitor [104], indicating that inhibition of PDGFR signaling has therapeutic benefits in AIDS-KS and strengthens the idea that PDGFRA can be an oncogenic driver in AIDS-KS. In murine KS, sunitinib, a drug approved for renal cell carcinoma and tested in Phase 1 clinical trials for tumors in the context of HIV/AIDS [105], had a more potent antitumor effect than imatinib, leading to complete inhibition of tumor growth, which occurred with complete blockage of PDGFRA phosphorylation [33]. However, both imatinib and sunitinib inhibit a number of other RTKs, including KIT and VEGFR, which are also expressed and thought to be relevant in KS [29,62], so it is difficult to know if the clinical responses are solely due to inhibition of PDGFRA. Nevertheless, these results suggest that therapeutic approaches to complete inhibition of PDGFRA signaling can be highly efficacious in KS treatment (see Clinician's corner). More importantly, it strengthens the case for a circulating PDGFRA-expressing MSC precursor in the ontogeny of KS.

Concluding remarks

Evidence emanating from different experimental models of *de novo* KSHV-infected progenitors allows the development of an emergent picture of the origin of the KS spindle oncogenic progenitor cell. Circulating progenitor cells can serve as KS oncogenic progenitors recruited by inflammatory and KS-secreted cytokines, inducing spindle cell reprogramming and transformation upon KSHV infection. This process explains the propensity of KS lesions to localize to scar tissue or sites of inflammation, growing in a cytokine-rich microenvironment. These circulating progenitor cells can be either from the endothelial or mesenchymal lineage, since KSHV can reprogram one into the other through EndMT and/or MEndT. This suggests that both cell types are probably critical at different stages of the oncogenic process through the endothelial–mesenchymal differentiation axis induced by KSHV. Sarcomas are tumors of mesenchymal cell origin; KS is not the exception, as most KS lesions express the MSC-defining marker PDGFRA. As the lesions and clinical studies suggest, the final steps toward oncogenic transformation by KSHV probably involve PDGFRA-positive progenitor cells through a KSHV-induced ligand-mediated activation of the PDGFRA signaling-driven mechanism of oncogenesis. KSHV biology suggests that this can happen via the infection of ECs, likely of lymphatic origin leading to EndMT, or through infection of recruited circulating or resident MSCs leading to MEndT with an expression of both endothelial markers such as VEGFR2 and R3 and mesenchymal markers such as PDGFRA and vimentin,

Outstanding questions

Which cell lineages are the KS progenitor cells: mesenchymal or endothelial circulating progenitor cells, or both?

Are both cell lineages essential for KS development? Is the interaction between these cell lineages important for the KS spindle cell progenitors to achieve the hallmarks of cancer?

How important are MEndT/EndMT in virus-induced oncogenic processes?

Are these transitions new targets for discovering therapies for KS?

How would complete blockage of RTK signaling affect the therapeutic benefits of RTK inhibitors in AIDS-KS, and would specific inhibition of PFGFRA, be sufficient or is inhibition of multiple kinases more effective?

characteristic of the KS spindle cell (see Outstanding questions). The unique ability of KSHV for reprogramming ECs and MSCs provides new insights into the study of the mesenchymal–endothelial differentiation axis and the importance of this mechanism in the process of cancer and viral oncogenesis. Moreover, this knowledge opens the door for testing strategies to inhibit this differentiation axis as therapeutic targets for KS.

Acknowledgments

In memoriam of Dr. Enrique A. Mesri, an inspiring scientist, a great mentor, and friend, who sadly passed away days before sending the manuscript. For his dedication and contributions to the KSHV and viral oncology field, we will be always in debt to him.

Declaration of interests

None are declared by the authors.

References

1. Thiery, J.P. (2002) Epithelial-mesenchymal transitions in tumour progression. *Nat. Rev. Cancer* 2, 442–454
2. Kalluri, R. and Weinberg, R.A. (2009) The basics of epithelial-mesenchymal transition. *J. Clin. Invest.* 119, 1420–1428
3. Potenta, S. *et al.* (2008) The role of endothelial-to-mesenchymal transition in cancer progression. *Br. J. Cancer* 99, 1375–1379
4. Zeisberg, E.M. *et al.* (2007) Discovery of endothelial to mesenchymal transition as a source for carcinoma-associated fibroblasts. *Cancer Res.* 67, 10123–10128
5. Lovisa, S. *et al.* (2020) Endothelial-to-mesenchymal transition compromises vascular integrity to induce Myc-mediated metabolic reprogramming in kidney fibrosis. *Sci. Signal.* 13, eaaz25975
6. Ubil, E. *et al.* (2014) Mesenchymal-endothelial transition contributes to cardiac neovascularization. *Nature* 514, 585–590
7. Dong, W. *et al.* (2020) Mesenchymal-endothelial transition-derived cells as a potential new regulatory target for cardiac hypertrophy. *Sci. Rep.* 10, 6652
8. Cai, W. *et al.* (2021) PERK-eIF2alpha-ERK1/2 axis drives mesenchymal-endothelial transition of cancer-associated fibroblasts in pancreatic cancer. *Cancer Lett.* 515, 86–95
9. Batlle, R. *et al.* (2019) Regulation of tumor angiogenesis and mesenchymal-endothelial transition by p38alpha through TGF-beta and JNK signaling. *Nat. Commun.* 10, 3071
10. Medici, D. and Kalluri, R. (2012) Endothelial-mesenchymal transition and its contribution to the emergence of stem cell phenotype. *Semin. Cancer Biol.* 22, 379–384
11. Arciniegas, E. *et al.* (1992) Transforming growth factor beta 1 promotes the differentiation of endothelial cells into smooth muscle-like cells *in vitro*. *J. Cell Sci.* 103, 521–529
12. Piera-Velazquez, S. and Jimenez, S.A. (2019) Endothelial to mesenchymal transition: role in physiology and in the pathogenesis of human diseases. *Physiol. Rev.* 99, 1281–1324
13. Evrard, S.M. *et al.* (2016) Endothelial to mesenchymal transition is common in atherosclerotic lesions and is associated with plaque instability. *Nat. Commun.* 7, 11853
14. Chen, L. *et al.* (2021) Vitamin D3 induces mesenchymal-to-endothelial transition and promotes a proangiogenic niche through IGF-1 signaling. *iScience* 24, 102272
15. Gurzu, S. *et al.* (2013) Mesenchymal-to-endothelial transition in Kaposi sarcoma: a histogenetic hypothesis based on a case series and literature review. *PLoS One* 8, e71530
16. Chang, Y. *et al.* (1994) Identification of herpesvirus-like DNA sequences in AIDS-associated Kaposi's sarcoma. *Science* 266, 1865–1869
17. Gramolelli, S. and Ojala, P.M. (2017) Kaposi's sarcoma herpesvirus-induced endothelial cell reprogramming supports viral persistence and contributes to Kaposi's sarcoma tumorigenesis. *Curr. Opin. Virol.* 26, 156–162
18. Cancian, L. *et al.* (2013) Cellular origin of Kaposi's sarcoma and Kaposi's sarcoma-associated herpesvirus-induced cell reprogramming. *Trends Cell Biol.* 23, 421–432
19. Ganem, D. (2010) KSHV and the pathogenesis of Kaposi sarcoma: listening to human biology and medicine. *J. Clin. Invest.* 120, 939–949
20. Salahuddin, S.Z. *et al.* (1988) Angiogenic properties of Kaposi's sarcoma-derived cells after long-term culture *in vitro*. *Science* 242, 430–433
21. Samaniego, F. *et al.* (1995) Inflammatory cytokines induce AIDS-Kaposi's sarcoma-derived spindle cells to produce and release basic fibroblast growth factor and enhance Kaposi's sarcoma-like lesion formation in nude mice. *J. Immunol.* 154, 3582–3592
22. Ensoli, B. *et al.* (1989) AIDS-Kaposi's sarcoma-derived cells express cytokines with autocrine and paracrine growth effects. *Science* 243, 223–226
23. Browning, P.J. *et al.* (1994) Identification and culture of Kaposi's sarcoma-like spindle cells from the peripheral blood of human immunodeficiency virus-1-infected individuals and normal controls. *Blood* 84, 2711–2720
24. Cesarman, E. *et al.* (2019) Kaposi sarcoma. *Nat. Rev. Dis. Primers* 5, 9
25. Casper, C. (2011) The increasing burden of HIV-associated malignancies in resource-limited regions. *Annu. Rev. Med.* 62, 157–170
26. Sullivan, R.J. *et al.* (2008) HIV/AIDS: epidemiology, pathophysiology, and treatment of Kaposi sarcoma-associated herpesvirus disease: Kaposi sarcoma, primary effusion lymphoma, and multicentric Castleman disease. *Clin. Infect. Dis.* 47, 1209–1215
27. Nguyen, H.Q. *et al.* (2008) Persistent Kaposi sarcoma in the era of highly active antiretroviral therapy: characterizing the predictors of clinical response. *AIDS* 22, 937–945
28. Schulz, T.F. and Cesarman, E. (2015) Kaposi sarcoma-associated herpesvirus: mechanisms of oncogenesis. *Curr. Opin. Virol.* 14, 116–128
29. Mesri, E.A. *et al.* (2010) Kaposi's sarcoma and its associated herpesvirus. *Nat. Rev. Cancer* 10, 707–719
30. Mocroft, A. *et al.* (2004) The changing pattern of Kaposi sarcoma in patients with HIV, 1994-2003: the EuroSIDA Study. *Cancer* 100, 2644–2654
31. Glesby, M.J. *et al.* (1996) Use of antiherpes drugs and the risk of Kaposi's sarcoma: data from the Multicenter AIDS Cohort Study. *J. Infect. Dis.* 173, 1477–1480
32. Mutlu, A.D. *et al.* (2007) *In vivo*-restricted and reversible malignancy induced by human herpesvirus-8 KSHV: a cell and animal model of virally induced Kaposi's sarcoma. *Cancer Cell* 11, 245–258
33. Cavallin, L.E. *et al.* (2018) KSHV-induced ligand mediated activation of PDGF receptor-alpha drives Kaposi's sarcomagenesis. *PLoS Pathog.* 14, e1007175
34. Wang, L. *et al.* (2006) Immortalization of primary endothelial cells by the K1 protein of Kaposi's sarcoma-associated herpesvirus. *Cancer Res.* 66, 3658–3666

35. Naipauer, J. *et al.* (2019) PDGFRA defines the mesenchymal stem cell Kaposi's sarcoma progenitors by enabling KSHV oncogenesis in an angiogenic environment. *PLoS Pathog.* 15, e1008221
36. Muralidhar, S. *et al.* (1998) Identification of kaposin (open reading frame K12) as a human herpesvirus 8 (Kaposi's sarcoma-associated herpesvirus) transforming gene. *J. Virol.* 72, 4980–4988
37. Cavallin, L.E. *et al.* (2014) Molecular and cellular mechanisms of KSHV oncogenesis of Kaposi's sarcoma associated with HIV/AIDS. *PLoS Pathog.* 10, e1004154
38. Montaner, S. *et al.* (2003) Endothelial infection with KSHV genes *in vivo* reveals that vGPCR initiates Kaposi's sarcomagenesis and can promote the tumorigenic potential of viral latent genes. *Cancer Cell* 3, 23–36
39. Chang, H.H. and Ganem, D. (2013) A unique herpesviral transcriptional program in KSHV-infected lymphatic endothelial cells leads to mTORC1 activation and rapamycin sensitivity. *Cell Host Microbe* 13, 429–440
40. Montaner, S. *et al.* (2006) The Kaposi's sarcoma-associated herpesvirus G protein-coupled receptor as a therapeutic target for the treatment of Kaposi's sarcoma. *Cancer Res.* 66, 168–174
41. Bais, C. *et al.* (2003) Kaposi's sarcoma associated herpesvirus G protein-coupled receptor immortalizes human endothelial cells by activation of the VEGF receptor-2/ KDR. *Cancer Cell* 3, 131–143
42. Mesri, E.A. *et al.* (2014) Human viral oncogenesis: a cancer hallmarks analysis. *Cell Host Microbe* 15, 266–282
43. Wang, H.W. *et al.* (2004) Kaposi sarcoma herpesvirus-induced cellular reprogramming contributes to the lymphatic endothelial gene expression in Kaposi sarcoma. *Nat. Genet.* 36, 687–693
44. Hong, Y.K. *et al.* (2004) Lymphatic reprogramming of blood vascular endothelium by Kaposi sarcoma-associated herpesvirus. *Nat. Genet.* 36, 683–685
45. Jussila, L. *et al.* (1998) Lymphatic endothelium and Kaposi's sarcoma spindle cells detected by antibodies against the vascular endothelial growth factor receptor-3. *Cancer Res.* 58, 1599–1604
46. Regezi, J.A. *et al.* (1993) Human immunodeficiency virus-associated oral Kaposi's sarcoma. A heterogeneous cell population dominated by spindle-shaped endothelial cells. *Am. J. Pathol.* 143, 240–249
47. Weich, H.A. *et al.* (1991) AIDS-associated Kaposi's sarcoma-derived cells in long-term culture express and synthesize smooth muscle alpha-actin. *Am. J. Pathol.* 139, 1251–1258
48. Nakamura, S. *et al.* (1988) Kaposi's sarcoma cells: long-term culture with growth factor from retrovirus-infected CD4+ T cells. *Science* 242, 426–430
49. Flore, O. *et al.* (1998) Transformation of primary human endothelial cells by Kaposi's sarcoma-associated herpesvirus. *Nature* 394, 588–592
50. Ciufo, D.M. *et al.* (2001) Spindle cell conversion by Kaposi's sarcoma-associated herpesvirus: formation of colonies and plaques with mixed lytic and latent gene expression in infected primary dermal microvascular endothelial cell cultures. *J. Virol.* 75, 5614–5626
51. Ojala, P.M. and Schulz, T.F. (2014) Manipulation of endothelial cells by KSHV: implications for angiogenesis and aberrant vascular differentiation. *Semin. Cancer Biol.* 26, 69–77
52. Krug, L.T. *et al.* (2004) Inhibition of infection and replication of human herpesvirus 8 in microvascular endothelial cells by alpha interferon and phosphonoformic acid. *J. Virol.* 78, 8359–8371
53. Grundhoff, A. and Ganem, D. (2004) Inefficient establishment of KSHV latency suggests an additional role for continued lytic replication in Kaposi sarcoma pathogenesis. *J. Clin. Invest.* 113, 124–136
54. Renne, R. *et al.* (1998) Limited transmission of Kaposi's sarcoma-associated herpesvirus in cultured cells. *J. Virol.* 72, 5182–5188
55. Moses, A.V. *et al.* (1999) Long-term infection and transformation of dermal microvascular endothelial cells by human herpesvirus 8. *J. Virol.* 73, 6892–6902
56. Lipps, C. *et al.* (2017) Proliferation status defines functional properties of endothelial cells. *Cell. Mol. Life Sci.* 74, 1319–1333
57. Dubich, T. *et al.* (2021) 3D culture conditions support Kaposi's sarcoma herpesvirus (KSHV) maintenance and viral spread in endothelial cells. *J. Mol. Med. (Berl.)* 99, 425–438
58. Dubich, T. *et al.* (2019) An endothelial cell line infected by Kaposi's sarcoma-associated herpes virus (KSHV) allows the investigation of Kaposi's sarcoma and the validation of novel viral inhibitors *in vitro* and *in vivo*. *J. Mol. Med. (Berl.)* 97, 311–324
59. Lagunoff, M. *et al.* (2002) *De novo* infection and serial transmission of Kaposi's sarcoma-associated herpesvirus in cultured endothelial cells. *J. Virol.* 76, 2440–2448
60. Roy, D. *et al.* (2013) mTOR inhibitors block Kaposi sarcoma growth by inhibiting essential autocrine growth factors and tumor angiogenesis. *Cancer Res.* 73, 2235–2246
61. An, F.Q. *et al.* (2006) Long-term-infected telomerase-immortalized endothelial cells: a model for Kaposi's sarcoma-associated herpesvirus latency *in vitro* and *in vivo*. *J. Virol.* 80, 4833–4846
62. Moses, A.V. *et al.* (2002) Kaposi's sarcoma-associated herpesvirus-induced upregulation of the c-kit proto-oncogene, as identified by gene expression profiling, is essential for the transformation of endothelial cells. *J. Virol.* 76, 8383–8399
63. Raggo, C. *et al.* (2005) Novel cellular genes essential for transformation of endothelial cells by Kaposi's sarcoma-associated herpesvirus. *Cancer Res.* 65, 5084–5095
64. McAllister, S.C. *et al.* (2004) Kaposi sarcoma-associated herpesvirus (KSHV) induces heme oxygenase-1 expression and activity in KSHV-infected endothelial cells. *Blood* 103, 3465–3473
65. Rose, P.P. *et al.* (2007) Insulin-like growth factor II receptor-mediated intracellular retention of cathepsin B is essential for transformation of endothelial cells by Kaposi's sarcoma-associated herpesvirus. *J. Virol.* 81, 8050–8062
66. Gay, L.A. *et al.* (2021) Kaposi's sarcoma-associated herpesvirus microRNA mutants modulate cancer hallmark phenotypic differences in human endothelial cells. *J. Virol.* 95, e02022-20
67. Sethuraman, S. *et al.* (2017) microRNA dependent and independent deregulation of long non-coding RNAs by an oncogenic herpesvirus. *PLoS Pathog.* 13, e1006508
68. Dai, L. *et al.* (2017) ABC294640, A novel sphingosine kinase 2 inhibitor, induces oncogenic virus-infected cell autophagic death and represses tumor growth. *Mol. Cancer Ther.* 16, 2724–2734
69. Yu, X. *et al.* (2016) Suppression of KSHV-induced angiopoietin-2 inhibits angiogenesis, infiltration of inflammatory cells, and tumor growth. *Cell Cycle* 15, 2053–2065
70. Ye, F. *et al.* (2012) Nutlin-3 induces apoptosis, disrupts viral latency and inhibits expression of angiopoietin-2 in Kaposi sarcoma tumor cells. *Cell Cycle* 11, 1393–1399
71. Beauclair, G. *et al.* (2020) Targeting Kaposi's sarcoma-associated herpesvirus ORF21 tyrosine kinase and viral lytic reactivation by tyrosine kinase inhibitors approved for clinical use. *J. Virol.* 94, e01791-19
72. Golas, G. *et al.* (2019) Characterization of *de novo* lytic infection of dermal lymphatic microvascular endothelial cells by Kaposi's sarcoma-associated herpesvirus. *Virology* 536, 27–31
73. Carroll, P.A. *et al.* (2004) Kaposi's sarcoma-associated herpesvirus infection of blood endothelial cells induces lymphatic differentiation. *Virology* 328, 7–18
74. Yoo, J. *et al.* (2012) Opposing regulation of PROX1 by interleukin-3 receptor and NOTCH directs differential host cell fate reprogramming by Kaposi sarcoma herpes virus. *PLoS Pathog.* 8, e1002770
75. Choi, D. *et al.* (2020) The lymphatic cell environment promotes kaposi sarcoma development by prox1-enhanced productive lytic replication of Kaposi sarcoma herpes virus. *Cancer Res.* 80, 3130–3144
76. Gramolelli, S. *et al.* (2020) Oncogenic herpesvirus engages endothelial transcription factors SOX18 and PROX1 to increase viral genome copies and virus production. *Cancer Res.* 80, 3116–3129

77. Gasperini, P. *et al.* (2012) Kaposi sarcoma herpesvirus promotes endothelial-to-mesenchymal transition through Notch-dependent signaling. *Cancer Res.* 72, 1157–1169
78. Cheng, F. *et al.* (2011) KSHV-initiated notch activation leads to membrane-type-1 matrix metalloproteinase-dependent lymphatic endothelial-to-mesenchymal transition. *Cell Host Microbe* 10, 577–590
79. Mesri, E.A. and Cesarman, E. (2011) Kaposi's sarcoma herpesvirus oncogenesis is a notch better in 3D. *Cell Host Microbe* 10, 529–531
80. Webster-Cyriaque, J. (2002) Development of Kaposi's sarcoma in a surgical wound. *N. Engl. J. Med.* 346, 1207–1210
81. Taddeo, A. *et al.* (2008) Circulating endothelial progenitor cells are increased in patients with classic Kaposi's sarcoma. *J. Invest. Dermatol.* 128, 2125–2128
82. Della Bella, S. *et al.* (2008) Peripheral blood endothelial progenitors as potential reservoirs of Kaposi's sarcoma-associated herpesvirus. *PLoS One* 3, e1520
83. Barozzi, P. *et al.* (2003) Post-transplant Kaposi sarcoma originates from the seeding of donor-derived progenitors. *Nat. Med.* 9, 554–561
84. Ashlock, B.M. *et al.* (2014) Productively infected murine Kaposi's sarcoma-like tumors define new animal models for studying and targeting KSHV oncogenesis and replication. *PLoS One* 9, e87324
85. Jones, T. *et al.* (2012) Direct and efficient cellular transformation of primary rat mesenchymal precursor cells by KSHV. *J. Clin. Invest.* 122, 1076–1081
86. Moonen, J.R. *et al.* (2010) Endothelial progenitor cells give rise to pro-angiogenic smooth muscle-like progeny. *Cardiovasc. Res.* 86, 506–515
87. Morikawa, S. *et al.* (2009) Prospective identification, isolation, and systemic transplantation of multipotent mesenchymal stem cells in murine bone marrow. *J. Exp. Med.* 206, 2483–2496
88. Rodriguez, R. *et al.* (2012) Modeling sarcomagenesis using multipotent mesenchymal stem cells. *Cell Res.* 22, 62–77
89. Parsons, C.H. *et al.* (2004) Susceptibility of human fetal mesenchymal stem cells to Kaposi sarcoma-associated herpesvirus. *Blood* 104, 2736–2738
90. Yoo, S.M. *et al.* (2014) Kaposi's sarcoma-associated herpesvirus infection of human bone-marrow-derived mesenchymal stem cells and their angiogenic potential. *Arch. Virol.* 159, 2377–2386
91. Lee, M.S. *et al.* (2016) Human mesenchymal stem cells of diverse origins support persistent infection with Kaposi's sarcoma-associated herpesvirus and manifest distinct angiogenic, invasive, and transforming phenotypes. *MBio* 7, e02109–15
92. Li, Y. *et al.* (2018) Evidence for Kaposi sarcoma originating from mesenchymal stem cell through KSHV-induced mesenchymal-to-endothelial transition. *Cancer Res.* 78, 230–245
93. Wang, X. *et al.* (2020) KSHV enhances mesenchymal stem cell homing and promotes KS-like pathogenesis. *Virology* 549, 5–12
94. Chen, Q. *et al.* (2021) Kaposi's sarcoma herpesvirus is associated with osteosarcoma in Xinjiang populations. *Proc. Natl. Acad. Sci. U. S. A.* 118, e2016653118
95. Ding, Y. *et al.* (2021) Kaposi's sarcoma-associated herpesvirus promotes mesenchymal-to-endothelial transition by resolving the bivalent chromatin of PROX1 gene. *PLoS Pathog.* 17, e1009847
96. Chen, W. *et al.* (2021) Kaposi's sarcoma-associated herpesvirus vFLIP promotes MEndT to generate hybrid M/E state for tumorigenesis. *PLoS Pathog.* 17, e1009600
97. Ensoli, B. *et al.* (2001) Biology of Kaposi's sarcoma. *Eur. J. Cancer* 37, 1251–1269
98. Sturzl, M. *et al.* (1992) Expression of platelet-derived growth factor and its receptor in AIDS-related Kaposi sarcoma in vivo suggests paracrine and autocrine mechanisms of tumor maintenance. *Proc. Natl. Acad. Sci. U. S. A.* 89, 7046–7050
99. Werner, S. *et al.* (1990) Cultured Kaposi's sarcoma-derived cells express functional PDGF A-type and B-type receptors. *Exp. Cell Res.* 187, 98–103
100. Roth, W.K. *et al.* (1989) Depletion of PDGF from serum inhibits growth of AIDS-related and sporadic Kaposi's sarcoma cells in culture. *Oncogene* 4, 483–487
101. Koster, R. *et al.* (1996) Consensus-interferon and platelet-derived growth factor adversely regulate proliferation and migration of Kaposi's sarcoma cells by control of c-myc expression. *Am. J. Pathol.* 149, 1871–1885
102. Corless, C.L. *et al.* (2011) Gastrointestinal stromal tumours: origin and molecular oncology. *Nat. Rev. Cancer* 11, 865–878
103. Ho, A.L. *et al.* (2012) PDGF receptor alpha is an alternative mediator of rapamycin-induced Akt activation: implications for combination targeted therapy of synovial sarcoma. *Cancer Res.* 72, 4515–4525
104. Koon, H.B. *et al.* (2014) Phase II trial of imatinib in AIDS-associated Kaposi's sarcoma: AIDS Malignancy Consortium Protocol 042. *J. Clin. Oncol.* 32, 402–408
105. Rudek, M.A. *et al.* (2014) A phase 1/pharmacokinetic study of sunitinib in combination with highly active antiretroviral therapy in human immunodeficiency virus-positive patients with cancer: AIDS Malignancy Consortium trial AMC 061. *Cancer* 120, 1194–1202