Kaposi Sarcoma with Abscessing Clinicopathologic Morphology

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Abstract

Background: The AIDS epidemic heralded an expanded spectrum of Kaposi sarcoma (KS) variants but to date KS masquerading as an abscess on clinical and histopathological assessment is undocumented.

Methods: This 3 year retrospective study re-appraised the clinicopathological profile of all biopsies of KS that presented as abscesses clinically and demonstrated abscessing or suppurrative microscopic features.

Results: Ten males and 9 females form the study cohort. The clinical diagnosis in 11 patients was exclusively that of an abscess. Tuberculosis and abscess were clinical differential diagnoses in 8 patients. The average CD4 count was 261 cells/mm³. Abscessing KS was a sentinel of HIV infection and AIDS in 4 patients. Of 11 patients on HAART, 3 were virally suppressed. The pathology appeared within 3 months of HAART commencement in 5 patients; repeat CD4 counts and viral load assessments were not available for scrutiny. Microscopically, 17/19 initial biopsies with abscessing morphology demonstrated typical features of KS at least focally, while 2/19 had an exclusive abscess-like morphology. Spindle cell heterogeneity was present with co-existence in all biopsies of myofibroblasts and malignant spindle cells, the latter were HHV8-LANA-1 positive. Special stains and polymerase chain reaction investigation for infections were negative and were crucial in excluding an infective cause. Sequestrum was noted focally in the exudate of one biopsy but the patient’s demise precluded assessment of underlying osteitis. Folliculitis was not present.

Conclusion: HHV8-LANA-1 immunostaining underpins the identification of KS that masquerades as an abscess clinically and may mimic a spectrum of infections microscopically. Heightened awareness of this inflammatory response is pivotal to its diagnosis. The exact cause of the abscessing morphology was unconfirmed in the present study, but on-going investigation is critical to determine the pathogenesis of this, hitherto undescribed, clinicopathological profile of KS, that may predicate altered therapeutic approaches.

Keywords: Kaposi sarcoma; Suppuration; Pyogenic; Abscess; HIV; AIDS

Introduction

Kaposi sarcoma (KS), a multicentric angioproliferative condition [1], is associated with Human Herpes Virus 8 (HHV8) infection and widespread anatomical involvement, including cutaneous, mucosal and internal visceral sites. Described originally by Mortiz Kaposi over a century ago [2], it is categorized into 4 clinico-epidemiological forms: classic, African (endemic), AIDS-associated (epidemic) and iatrogenic KS subtypes. KS is regarded as a low grade tumor but an aggressive course occurs in >50% of patients in the AIDS context [3]. Irrespective of the clinical subgroup of KS, similar cutaneous histopathological features occur for well-known KS variants that encompass 3 distinctive morphological patch, plaque and nodular stages with potential for morphological overlap. The HIV/AIDS era, however, has introduced an expanded spectrum of histomorphological variants [4-6]. Furthermore, the microscopic features of KS lesions are impacted by co-morbid, concomitant immunosuppressive diseases, and anti-retroviral drug-induced flares and regression [2,7-11]. Hence, the histomorphological spectrum of KS is currently characterized by well-known established, uncommon and emerging variants [12]. Notwithstanding these emerging variants, to date, clinically unsuspected KS lesions masquerading as abscesses, with histopathological features of KS masked by exuberant suppurrative inflammation, justifying an appellation of ‘abscessing’ Kaposi sarcoma, is a hitherto unreported phenomenon, with potential serious patient management implications.

This study was therefore undertaken to assess the clinicopathological features of KS with abscessing morphology and to highlight potential diagnostic mimicry and pitfalls. In addition, potential pathogenetic mechanisms for the abscessing presentation are discussed.

Materials and Methods

This is a descriptive 3 year retrospective study that involved a re-appraisal of all biopsies of KS with an abscessing morphology that were diagnosed in the Department of Anatomical Pathology, National Health Laboratory Service, Durban, KwaZulu-Natal from 1 January 2012 to 31 December 2014. The SNOMED word search engine targeting ‘Kaposi’ and ‘abscess’ was employed to identify confirmed biopsies of KS with abscessing morphology for study purposes. The clinical details that were obtained from the histopathology request forms included age, sex, race, presenting symptoms, laterality, imaging findings, HIV status, CD4 counts, HIV viral load, clinical differential diagnosis, exposure to highly active anti-retroviral therapy (HAART) and the interval between HAART initiation and...
disease onset. The histopathology report provided information on gross characteristics of the submitted specimen.

Using the histopathology accession numbers, slides were retrieved from the archive of the Department of Anatomical Pathology/National Health Laboratory Service, Durban, KwaZulu-Natal for re-evaluation. Sections of biopsies that had faded were recut and stained with hematoxylin and eosin to assess the presence of chronic supplicative inflammation typical of abscess wall, morphologic subtype of KS, ulceration, sequestrum and bacterial colonies. Ziehl Neelsen, periodic acid Schiff, Warthin Starry and Brown Brenn Hopps stains were undertaken to identify potential intra-lesional mycobacteria, fungi, Bartonella species and Gram-positive and Gram-negative bacteria, respectively. Immunohistochemical immunoprofiling for HHV8-LANA-1 (Novocasta, Newcastle-Upon-Tyne, UK, clone3B10, 1:40 dilution) and endothelial cells using anti-CD34 antibody (DakoCytomation, Glostrup, Denmark, clone QBEND-10, 1:40 dilution) and anti-CD31 antibody (DakoCytomation, Glostrup, Denmark, clone JC70, 1:30 dilution) were undertaken on 2 µm thick sections cut from archival wax blocks. The tissue sections were subjected to heat-assisted microwave antigen retrieval and manual polymer-based immunohistochemical staining with diaminobenzidine as the chromogen, using a standard immunohistochemical protocol with positive and negative controls. Genomic DNA was extracted from formalin-fixed paraffin-embedded tissues using the QIAamp DNA formalin-fixed tissue (Qiagen, Hilden, Germany) according to the manufacturer's protocol. Quantified DNA quality was thereafter assessed by PCR amplification of the 159 bp product of the reference gene, β-actin [13]. PCR investigations were undertaken to detect *Mycobacterium tuberculosis* (MTb) and *Bartonella* species. Samples that were negative following agarose gel electrophoresis on the first outer PCR were then subjected to nested PCR. All PCR amplification was performed on a CFX96 Real-time PCR system (BIO-RAD, CA, USA). *Bartonella* species identification was accomplished by the double PCR of a 126-326 bp region of the internal transcribed spacer region of ribosomal RNA using the primers (16SrDNA F: CGTTTCTCTTTCTTCAGATGATG and 16SrDNA R: CTCTAACCACTAGCTACAAAG). MTb complex identification was accomplished by PCR targeting of a 220 bp segment of IS6110 using the primers O1: CGGAGCACCCCGCGGCAAAGCCCGGAGAG; O2: CATCGTGGAAGCGCCCGCGGACCGAGAT) and a 123 bp segment using inner primers (I1: CCTGCGAGCGTAGCCGCGG and I2: CTCGGCGACCGCACCCTCG). Permission to conduct the study was obtained from the University of KwaZulu-Natal Biomedical Research Ethics Committee (BE366/15).

**Results**

5813 biopsies of KS were confirmed in the study period. Of these, 28 (0.44%) patients with biopsies of KS with abscessing morphology were identified. Adequate clinical information of 19 patients was available; biopsies from these 19 patients form the study cohort.

**Clinical details**

There were 10 males and 9 females. The average patient age was 27.8 (range: 12-50) years. The patients presented with painful, expanding masses in the toe (10), thigh (4), trunk (1), arm (1), neck (1) and anus (1). The mass lesions were recognized as abscesses clinically and the walls thereof were biopsied during the incision and drainage procedure. The average CD4 count was 261 (range: 7-1002) cells/mm³. Abscessing KS was a sentinel of the HIV infection in patients 4, 10, 12 and 14 (Table 1). Patients 1, 6, 8, 12 and 18 were on anti-tuberculous therapy for pulmonary tuberculosis. Of 11 patients on HAART, viral immunosuppression was confirmed in 3 patients (1, 5, 11). The pathology appeared within 2 and 3 months of commencement of HAART in 3 and 2 patients, respectively. Patient 10 had recurrence of KS after chemoradiation. Of 4 patients (patients 1, 6, 12, 18) with lymphadenopathy, 2 had Castleman disease, confirmed in cervical and inguinal lymph node biopsies. The other 2 demonstrated reactive lymphoid hyperplasia and sinus histiocytosis.

The clinical diagnosis was exclusively that of an abscess in 11 patients. Tuberculosis and abscess were clinical differential diagnoses in 8 patients. KS was not a clinical consideration for any of the abscessing lesions, although disseminated cutaneous KS was noted in all patients and pleural and oral lesions in 1 and 4 patients each, respectively, on follow-up assessment.

**Pathological findings**

Seventeen patients had a single biopsy while 2 patients had 2 biopsies each. A total of 21 biopsies were reviewed for study purposes.

**Gross details**

All biopsies were of incisional wedge type and were noted to be hemorrhagic with cream and yellow necrotic foci, compatible with the clinical impression of granulating abscess wall and associated abscess contents.

**Microscopic details**

Of the 19 initial biopsies with abscessing morphology, 17 demonstrated typical features of KS at least focally while 2/19 (patients 5, 15) had an exclusive abscess-like morphology. The focal areas of KS in 17/19 biopsies demonstrated a solid arrangement of hyperchromatic spindle cells with intervening slit-like vascular channels (Figure 1A), hyaline globules, hemosiderin pigment, variable mitoses and HHV8-
LANA-1 (Figure 1B), CD34 (Figure 1C) and CD31 (Figure 1D) immunopositivity. The abscessing morphology that dominated these 17 biopsies, noted centrally or in the deep, subcutaneous aspect of the sample, demonstrated organizing, chronic suppurative inflammation in granulation tissue, typical of an abscess wall and contents (Figures 2A and 2B). However, the spindle cells within the granulation tissue were heterogeneous; some were normochromatic and resembled myofibroblasts while others were hyperchromatic (Figure 2C). This prompted the conduction of HHV8-LANA-1 and α-smooth muscle actin immunostaining on these biopsies, that confirmed the divergent spindle cell population (Figures 2D and 2E), composed of a dual population of malignant KS spindle cells (Figure 2D) and myofibroblasts in the admixed organizing granulation tissue response (Figure 2E). Second deeper and wider biopsies from the same tumor site in 2 patients that had exclusive abscess-like morphology in the initial biopsy confirmed typical HHV8-LANA-1 positive KS.

The sarcomatous component from patients 4, 10, 12, 13 and 18 who had HAART exposure for 2 to 3 months, also demonstrated an infiltrate of lymphoid cells, neutrophils and histiocytes (Figures 3A and 3B). Eight biopsies demonstrated variable epidermal ulceration and non-specific, superficial bacterial colonization of the inflammatory exudate, but there was no connection between the ulcerated component and the deeper abscessing morphology (Figures 3C and 3D). Tuberculosis and bacillary angiomatosis were excluded in all biopsies by the lack of organisms on special infective stains. In addition PCR investigation for tuberculosis and bacillary angiomatosis were negative in all samples (Figures 4A and 4D). Focal sequestrum was identified within the abscessing component (patient 4) but attempts to confirm connection of the abscessing tumor with underlying osteitis on clinical review and imaging studies failed because the patient died shortly after biopsy. Folliculitis was not present in any biopsy.

![Figure 1: Nodular Kaposi sarcoma (A) composed of hyperchromatic spindle cells and intervening slit-like vascular channels (asterisks) (H&E, 240X) demonstrating brown nuclear HHV8-LANA-1 (B) (480X) and brown cytoplasmic CD34 (C) (480X) and CD31 (D) (480X) immunopositivity.](image1)

![Figure 2: Abscessing morphology with (A) chronic suppurative inflammation in granulation tissue (white asterisks) and central breakdown with necrotic and karyorrhectic debris (black asterisk) (H&E, 120X). High power demonstration (B) of vasculature (black asterisk) in granulation tissue and suppurative inflammation (white asterisk) (H&E, 240X). High power demonstration of hyperchromatic spindle cells (C, arrows) (H&E, 240X), HHV8-LANA-1 positive spindle cells (D, arrows) (480X) and α-smooth muscle actin immunopositive myofibroblasts in granulation tissue (E, arrows) (480X).](image2)

![Figure 3: Biopsy from patient 9 in third month of anti-retroviral therapy demonstrating inflamed granulation tissue with capillaries (A, arrows) and neutrophils (A, asterisks) (H&E, 120X) and HHV8-LANA-1 positive cells therein (B, arrows). Acute inflammatory exudate (C, white asterisks) on the superficial aspect of ulcerated Kaposi sarcoma (C, black asterisks) with scattered HHV8-LANA-1 positive spindle cells (D, arrows).](image3)
The emergence of new KS variants in the AIDS era has not only expanded the histopathological spectrum but also introduced a plethora of mimickers and diagnostic pitfalls in the wake of expanding KS to AIDS-related and epidemic KS. Historically, KS has been associated with clinical and histopathological heterogeneity, although the latter is not unique to any specific clinical disease stage or presentation. The emergence of new KS variants in the AIDS era has not only expanded the histopathological spectrum but also introduced a plethora of mimickers and diagnostic pitfalls in its wake [12,15].

Currently, the histopathological diagnostic challenges include those related to mimicry of common well-recognized, uncommon poorly-recognized and newly-recognized KS variants, procedure and treatment-related changes and the impact of co-morbid co-lesional pathology on KS lesions [16]. Apart from heightened awareness and recognition of the changing face of KS in the AIDS era, diagnostic confirmation has also been facilitated by the recent discovery of Human herpesvirus-8 (HHV8), also labelled KS-associated herpes virus, as the viral etiopathogenesis of KS [17]. This has facilitated improved understanding of KS and therapeutic possibilities and distinction from diagnostic masqueraders [17-23]. The commercial availability of HHV-8 LANA-1 immunohistochemical antibodies and HHV8 PCR studies has enabled the diagnostic confirmation of KS in challenging histodiagnostic settings [19-22]. HHV-8 LANA-1 is a nuclear immunohistochemical antigen that is expressed by all cells that are latently infected by the virus. It is more reliable than PCR investigation as it demonstrates the nuclear immunolocalization of the viral product, in contrast to potential HHV-8 passenger lymphocytes and non-specific amplification, that may produce false positive results on PCR testing [20,21].

Because of the clinical diagnosis and the presence of granulation tissue with a neutrophil-rich component, abscess is a compelling microscopic diagnosis [24] and risky pitfall. However, in all biopsies, careful assessment revealed the presence of short spindle cell fascicles at the periphery of the suppurative component or admixed hyperchromatic spindle cells, which is not a usual feature of an abscess wall. Abscess wall histomorphology mainly that of inflamed granulation tissue is characterized by a random arrangement of myofibroblasts and vasculature. Immunophenotypically, the spindle cell component in abscess wall and in granulation tissue is of myofibroblastic lineage and will stain with myoid markers, including α-smooth muscle actin, muscle specific actin, calponin and variably with desmin [25]. In contrast, the spindle cells in KS lack a myoid immunophenotype, stain negatively with myoid immunomarkers but positively with the endothelial markers, CD31 and CD34; these endothelial markers do not stain myofibroblasts in granulation tissue [26]. Granulation tissue may assume a pseudosarcomatous histomorphology and mimic KS closely [25,26]. In this context, negative HHV8 immunostaining will distinguish KS from abscess wall.

In the context of AIDS, tuberculous and fungal infections are potential mimickers of KS with an abscessing morphology. Whilst the classic granulomatous response of tuberculosis sets it apart from KS, the anergic response in immunosuppressed patients lacks a granulomatous phenotype. The anergic tuberculous responses may be lymphohistiocytic, acellular or neutrophil-rich with abundant karyorrhexis [27-30]. Although documented more than 6 decades ago [31], the neutrophilic and karyorrhectic form of anergic TB has received more attention in the HIV and AIDS era. Granuloma-poor, karyorrhectic, mycobacteria-rich anergic responses have been associated with low CD4 counts and impaired cell-mediated and humoral immunity [27,28]. Heightened awareness of the anergic tuberculous responses underpin the conduct of mycobacterial stains for the identification of myriad acid fast bacilli, typical of this tuberculous response. However, a negative infective profile should not default to a diagnosis of abscess because of the acute inflammatory background. Heightened recognition of the abscessing morphology of KS also underpins the conduction of HHV8 stains, pivotal to KS diagnosis.

Bacillary angiomatosis, caused by Bartonella sp., is a known clinical and histopathological mimic of KS because of the clinical morphology and microscopic vessel-rich morphology [12,16]. Bacillary angiomatosis is typically neutrophil-rich; in such settings, abscessing KS may be misdiagnosed as bacillary angiomatosis, especially as both lesions stain positively with endothelial markers. However, HHV8-LANA-1 is negative in bacillary angiomatosis [12,16,21]. Hence, the gold standard in distinguishing abscessing KS from its mimickers is the heightened
The exact pathogenesis of abscessing KS remains enigmatic and speculative, but the hypothetical pathogenetic possibilities highlight, not only potential patient prognostication and management implications, but also foreground a new inflammatory paradigm of KS, that requires accelerated investigation for improved understanding. The possible pathogenetic mechanisms for the intra-tumoral neutrophilic influx are categorized into those related to local, systemic HAART-related [32-35] and systemic HAART-unrelated [36,37] mechanisms (Table 2), the latter based on responses caused by tumor-associated neutrophils (TANs) in a wide range of cancers [38-50]. In this study, the local mechanisms for intra-tumoral suppuration include origin of the supplicative component from acute folliculitis in hair-bearing skin, post-traumatic and foreign bodies, entrapment of superficial acute inflammatory exudate and underlying osteitis. In the present study, only lesional tissue was submitted for appraisal. Careful appraisal of larger excision samples, including of the adjacent skin, will enable assessment of folliculitis in paraxialal skin. A toe biopsy contained a splinter, albeit away from the supplicative component. The role of such traumatic insults and foreign bodies in the pathogenesis of the abscessing presentation requires ongoing appraisal. Although not confirmed in the present study, underlying osteitis, especially in destroyed toes, may be the origin of suppuration. Imaging studies and the identification of sequestrum in such biopsies will facilitate such a hypothesis. Finally, many biopsies were ulcerated. The abscessing component was not continuous with the surface, but deep dermal extension of superficial inflammatory exudate and subsequent entrainment thereof, following cicatrization of the surface connection is a plausible mechanism, especially in older lesions. Wedge biopsies were assessed in this study. Complete appraisal of excised ‘abscesses’ may be more helpful in confirming the evolution of such entrapment in serial tissue sections.

**Table 2:** Potential pathogenetic mechanisms of ‘abscessing’ Kaposi sarcoma.

<table>
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<tr>
<th>Mechanism</th>
<th>Example</th>
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<tbody>
<tr>
<td>Anti-tumor role</td>
<td>Tumor cell lysis by reactive oxygen species, natural killer cells, antibody-dependent cell-mediated cytotoxicity [36]</td>
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<tr>
<td>Anti-tumor role</td>
<td>Apoptosis of tumor cells by TRAIL, her-2 [36]</td>
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<tr>
<td>Anti-tumor role</td>
<td>Inhibition of neutrophil influx by MMP-8 [36]</td>
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<tr>
<td>Anti-tumor role</td>
<td>Suppression of specific CD8-T-cell-induced responses [36]</td>
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<tr>
<td>Anti-tumor role</td>
<td>Possible cytokine-induced tumor cell destruction by:</td>
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<tr>
<td>Anti-tumor role</td>
<td>Neutrophil extracellular traps [36]</td>
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<tr>
<td>Anti-tumor role</td>
<td>Interleukin-8 [37]</td>
</tr>
<tr>
<td>Anti-tumor role</td>
<td>Possible cytotoxic protein-induced tumor cell destruction by:</td>
</tr>
<tr>
<td>Anti-tumor role</td>
<td>T-cell intracytoplasmic antibody-1 [37]</td>
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**Key:** TANs: Tumor- Associated Neutrophils, TRAIL: Tumor-Necrosis Factor Related Apoptosis-Inducing Ligand
documented. The histomorphological features that have been briefly described include either that of the typical spindle cell reaction of KS or of an intra-tumor mononuclear inflammatory cell infiltrate, especially of CD3-positive T-lymphocytes [8,34,56,57]. While a neutrophil-rich immune response is described in cryptococal and tuberculous IRIS reactions, it is not documented in KS, to date [53,54]. In the present study, 5 patients manifested with expanding lesions within 2 to 3 months of initiation of HAART. While it is tempting to implicate an immune reconstitution basis for the abscessing morphology in these patients because of the temporal association with HAART exposure, follow-up CD4 counts and viral load assays, pivotal to the diagnosis of IRIS [33], were not traceable, to formalize a definitive hypothesis but require investigation in future studies.

TANs, initially regarded as bystander cells, are now a well-recognized component of the tumor milieu [36], and require consideration in KS, especially when local factors and KS-associated IRIS are not feasible pathogenetic options. The cytokines and chemokines in neutrophil granules may recruit additional cells and are capable of effecting protumoral and anti-tumor activity (Table 2). TANs have been identified in hematological (Hodgkin and non-Hodgkin anaplastic lymphomas [37,38], plasmacytoma [39], histiocytic sarcoma [40]), epithelial (esophageal [41], gastric [42], pancreaticobiliary [43,44], lung [45], head and neck squamous [36], breast [46,50], renal [47], urinary bladder carcinomas [48]) and germ cell [49] malignancies. In some settings, the neutrophil influx into tumors has been described as ‘lymphocytic’ [37], ‘supportive’ [60] or an ‘abscess’ [38-40,46] in clinical and histopathological presentations. Malignant tumors involving the lung [45], face [39] and bone [38] have masqueraded as an abscess clinically but to the best of our knowledge, KS masquerading as an abscess clinically has been documented only once to date, in a vulval location [61].

This study stands apart as the ‘abscessing’ KS clinical presentation was also recapitulated on microscopic assessment. KS is of endothelial origin and endothelial cells are pivotal in leukocyte recruitment and regulation thereof [62] through endothelial expression of selectin receptors and synthesis of chemokines and other chemotactic agents. It is therefore surprising that TANs are not part of the prototypical inflammatory response of KS [32]. Abrogation of leukocyte recruitment by lysosomal degradation of endothelial ICAM-1 and CD31 by HHV-8 has been hypothesized [63]. More recently, inhibition of neutrophilic migration by HHV8-infected endothelial cells through an interleukin-6 mechanism has also been proposed. Hence, while KS is apparently equipped with unique mechanisms that preclude the intra-tumor localization of neutrophils [62], the present study demonstrates breakdown of this property by virtue of the dense neutrophilic accumulation. Future investigation of the pathogenesis of KS with a clinicopathologic ‘abscessing’ morphology is mandatory to unravel the pathogenesis and prognostic role of this hitherto undocumented presentation, and to investigate potential innovative treatment tools.

Conclusion

KS with a neutrophil-rich infiltrate dense enough to assume clinical abscessing morphology is unrecognized to date, and even in high AIDS incidence regions such as ours, remains relatively rare. It is uncertain whether this is a truly rare presentation of KS or an under-recognized entity. Notwithstanding this, apparent appraisal of all abscesses and abscess wall samples from patients with HIV infection and AIDS and heightened recognition of abscessing KS underpin the correct diagnosis of this morphological presentation. KS is an AIDS-defining condition and management and staging of patients with AIDS are dependent on its recognition. It is mandatory that the unusual and emerging variants of KS be recognized by clinicians and pathologists to ensure optimal diagnosis and patient management, especially when the KS variant is the sentinel clue to HIV infection and AIDS. The documentation of ‘abscessing’ KS not only expands the clinicopathological profile of KS but also highlights associated diagnostic mimicry and pathogenetic hiatus of this enigmatic AIDS-defining malignancy, that require detailed prospective investigation.

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References
