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MORPHOLOGICAL CHARACTERISTICS OF TUMORS OF BONE, CARTILAGE, AND SOFT TISSUES, WITH CLINICOPATHOLOGICAL CORRELATES OF VEGF, KI-67, AND CD34

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ABSTRACT

Background: Bone, cartilaginous, and soft-tissue tumors show overlapping microscopic structures that complicate grading and risk stratification. Proliferation and angiogenesis markers may be complementary to morphology, yet integrated clinicopathological descriptions within these three anatomic compartments are reported inconsistently. Methods: A retrospective observational study on 168 histologically confirmed primary tumors (bone, cartilage, and soft tissue) seen through a tertiary referral pathology service between January 2019 – December 2023. The hematoxylin–eosin morphology was recorded using a standardized checklist (cellularity, pleomorphism, matrix quality, necrosis, mitotic activity, vascular pattern, and invasive front). Immunohistochemistry (IHC) analyses involved VEGF, Ki-67 (MIB-1), and CD34 (microvessel density, MVD). Clinicoradiologic variables and metastatic localization were abstracted from the records. Associations were evaluated using χ^2/Fisher's exact tests, ANOVA/Kruskal–Wallis, logistic regression for metastasis, and Kaplan–Meier analyses for survival. Results: Tumor distribution was bone 62/168 (36.9%), cartilage 48/168 (28.6%), soft tissue 58/168 (34.5%). High-grade morphology (necrosis ≥10%, marked pleomorphism, mitoses ≥10/10 HPF) was most common in osteosarcoma and pleomorphic soft-tissue sarcoma phenotypes. Median Ki-67 index increased stepwise from benign/intermediate to malignant lesions (6% → 18% → 34%, $p < 0.001$). VEGF overexpression (H-score ≥200) correlated with higher CD34-MVD ($r = 0.52$, $p < 0.001$), as well as lung-predominant metastasis in bone sarcomas. Within multivariable modeling, Ki-67 ≥25% (adjusted OR 3.1) as well as CD34-MVD ≥45 vessels/HPF (adjusted OR 2.6) independently predicted metastatic disease. Conclusion: Across bone, cartilage, and soft tissue tumors, morphologic aggressiveness aligns with a reproducible angiogenesis–proliferation signature (high VEGF, high Ki-67, high CD34-MVD) that refines risk assessment beyond histology alone. Standardized morphologic checklists paired with targeted IHC may improve grading confidence and metastatic risk prediction.

KEYWORDS: angiogenesis; proliferation index; microvessel density; VEGF; Ki-67; CD34; clinicopathological correlation; metastatic pattern; sarcoma pathology; immunohistochemistry

1. INTRODUCTION

Tumors born from bone, cartilage and soft tissue are a diagnostically challenging spectrum for the many entities that seem to share convergent morphologic themes—spindle-cell proliferation, variable matrix production, necrosis and hemorrhage—despite markedly different lineages and molecular drivers. Modern classification stresses morphology and immunophenotype and increasingly genetics, as defined in WHO 5th edition soft tissue and bone tumor classification framework [1]. However, the routine diagnostic task remains heavily dependent on histologic pattern recognition and grading algorithms to quantitatively assess the potential for metastasis and to advise surgical margins.

Prognosis in musculoskeletal oncology is a function of both, anatomic extent as well as biologic aggressiveness. Surgical staging protocols introduced to the treatment of musculoskeletal sarcoma are still highly relevant because they classify grade, spread of lesions at compartmental boundaries and metastasis, which are all signs of tumor vs. host interface and barrier anatomy [2]. Grading (in particular for soft-tissue sarcoma) widely adopted grading methods, such as French FNCLCC, focus on differentiation, necrosis and mitotic activity since the latter have been linked to tumour metastasis propensity and survival [3]. However, morphologic measurements could be ambiguous especially for cartilaginous tumors where focus of cellularity and nuclear atypia and sampling may be key or clinical activities do not necessarily resemble histologic grade.

Protein-specific markers for cell proliferation and cell angiogenesis constitute a biomedical adjunct to morphology. Ki-67, a proliferation-associated nuclear antigen that arises in cycling cells is an objective estimate of cellular growth fraction and has been evaluated in a total of sarcoma subtypes as a prognostic adjunct. Osteosarcoma summarizes our clinical evidence, finding stronger Ki-67 expression with advanced disease and metastasis and poorer overall survival [5]. Ki-67 has also shown prognostic utility in soft-tissue sarcoma cohorts, including synovial sarcoma [6]. Nonetheless, interpretation of Ki-67 can differ with hotspot selection and tumour heterogeneity, leading to demands for uniform assessment approaches.

Angiogenesis is also pivotal in tumor progression. As neovascularization is driven by VEGF (vascular endothelial growth factor), it promotes tumor growth and dissemination, and CD34 immunostaining allows determination of microvessel density (MVD) as a tissue-level surrogate for angiogenic function. Establishing a correlation between CD34-positive microvessels and tumorigenesis with VEGF expression in osteosarcoma through standard IHC, established a correlation between VEGF expression and adverse features including pulmonary metastasis [7]. Angiogenesis is also thought to be involved in aggressive soft-tissue sarcomas, where vascular morphology (e.g., “staghorn” vessels in solitary fibrous tumor) meets diagnostic morphology and, perhaps, clinical behavior [8].

In response, we developed a clinicopathological study that combined (i) a standardized morphologic checklist across bone, cartilage, and soft tissue tumors, and (ii) an IHC panel with targeting of VEGF, Ki-67, and CD34-MVD. We hypothesized that combining morphology and IHC profile was compatible with the metastatic localization and the shortterm prognosis compared to morphology alone.

WHO classification [1], musculoskeletal sarcoma staging [2], soft-tissue sarcoma grading [3], Ki-67 biology [4], Ki-67 in osteosarcoma prognosis [5], Ki-67 in synovial sarcoma prognosis [6], VEGF/CD34 in osteosarcoma prognosis [7], and vascular morphology diagnostic on soft tissue tumors [8].

2. MATERIALS AND METHODS

2.1. *Study design, setting, and duration*

A retrospective observational study was conducted in the Department of Pathology of a tertiary referral center. All eligible cases diagnosed between 01 January 2019 and 31 December 2023 were reviewed.

2.2. *Participants and case selection*

Consecutive cases were included if they met the following criteria:

Inclusion: (i) primary tumor arising in bone, cartilaginous tissue, or soft tissue; (ii) adequate formalin-fixed paraffin-embedded tissue for morphology and IHC; (iii) finalized diagnosis by musculoskeletal pathology consensus.

Exclusion: (i) post-therapy resection specimens without pre-treatment histology; (ii) metastatic tumors to bone/soft tissue from epithelial primaries; (iii) severely autolyzed/decacified tissue precluding reliable IHC interpretation.

2.3. *Ethics approval*

The study protocol was approved by the Institutional Ethics Committee (approval number recorded in departmental registry). Patient identifiers were removed before analysis.

2.4. *Morphological evaluation*

All slides were re-examined on hematoxylin–eosin staining. A standardized checklist recorded:

1. **Cellularity** (low/moderate/high)
2. **Cytologic atypia** (mild/moderate/marked)
3. **Mitotic activity** (mitoses per 10 high-power fields, HPF; hotspots)
4. **Tumor necrosis** (absent, <10%, 10–50%, >50%)
5. **Matrix pattern** (osteoid, chondroid, myxoid, collagenous, rhabdomyoblastic features)
6. **Growth pattern** (infiltrative vs pushing border)
7. **Vascular pattern** (thin-walled plexiform, staghorn-like, hyalinized vessels)
8. **Invasion** (cortex/medullary permeation; soft-tissue extension; perineural/vascular invasion when applicable)

Tumors were categorized into bone, cartilage, and soft-tissue groups, and subclassified by final histologic diagnosis.

2.5. Immunohistochemistry

IHC was performed on 3–4 µm sections using standardized antigen retrieval and detection.

- **Ki-67 (MIB-1):** reported as % nuclear positivity in 500 tumor cells in hotspots.
- **VEGF:** semi-quantitative H-score (0–300) based on intensity (0–3) × % area.
- **CD34:** microvessel density (MVD) counted in three vascular hotspots at 200×; mean vessels/HPF recorded. CD34 staining was interpreted for endothelial cells (not tumor cell CD34 expression) for MVD quantification.

2.6. Clinicopathological variables and outcome definitions

Age, sex, anatomic site, tumor size (cm), imaging extent, and metastasis localization (lung, lymph node, bone, other organs) were abstracted. Metastasis was defined by imaging and/or histologic confirmation during follow-up. Overall survival and metastasis-free survival were calculated from diagnosis date.

2.7. Statistical analysis

All analyses were performed using standard statistical software. Continuous variables were compared using ANOVA or Kruskal–Wallis. Categorical variables used χ^2 or Fisher's exact test. Correlations used Spearman coefficients. Logistic regression modeled metastatic disease. Survival was assessed by Kaplan–Meier with log-rank testing. A two-sided $p < 0.05$ was considered significant.

3. RESULTS

3.1. Narrative summary of key findings

In total, 168 tumors met inclusion criteria: bone (n=62), cartilage (n=48), and soft tissue (n=58). While the bone cohort was more often diagnosed with malignancy, the cartilage cohort consisted more of intermediate or low-grade lesions. The soft-tissue cases presented the greatest spectrum of morphologic diversity, ranging from relatively bland fibroblastic proliferations to high-grade pleomorphic sarcomas, with characteristic vascular patterns noted in selected entities. Across divisions was a common morphologic aggressiveness in a high-proliferation/high-angiogenesis phenotype. Tumors that showed necrosis ≥10% and mitotic activity ≥10/10 HPF exhibited significantly high Ki-67 indices and CD34-MVD. VEGF H-scores increased in parallel, leading to a predictable pattern: highest Ki-67 and VEGF levels were obtained in the most aggressive bone sarcomas and the most vigorous soft-tissue sarcomas, with the majority of cartilaginous tumors displaying intermediate Ki-67 values but wide variability according to grade and invasion pattern. Metastatic localization was consistent with musculoskeletal oncology of expected proportions with a lung predominance in high-grade bone sarcomas and select soft-tissue sarcomas. When tumors were stratified by marker thresholds (Ki-67 ≥25%, VEGF H-score ≥200, CD34-MVD ≥45 vessels/HPF), the probability of metastasis was markedly higher (independent of anatomic compartment).

Table 1: Baseline clinicopathological characteristics (n = 168)

Variable	Bone (n=62)	Cartilage (n=48)	Soft tissue (n=58)	p value
Age, mean ± SD (years)	21.8 ± 12.4	39.6 ± 15.1	42.3 ± 17.0	<0.001
Male sex, n (%)	38 (61.3)	28 (58.3)	33 (56.9)	0.89
Tumor size, median (IQR), cm	8.6 (6.2–12.4)	6.9 (4.5–9.2)	7.4 (4.8–10.6)	0.04
High grade morphology* n (%)	36 (58.1)	14 (29.2)	27 (46.6)	0.006
Metastasis during follow-up, n (%)	19 (30.6)	8 (16.7)	15 (25.9)	0.12
Lung metastasis (among metastatic), n (%)	15/19 (78.9)	5/8 (62.5)	10/15 (66.7)	—

Bone tumors also emerged at younger mean age with larger lesions based on the clinical characteristics of primary high-grade bone sarcomas. High-grade morphology was predominantly observed in bone and soft-tissue cohorts compared to cartilage tumors, indicating the high frequency of aggressive histotypes in these cohorts. Overall number of metastasis was not significantly different according to compartments, but lung metastasis was the predominant cancerous process in all three compartments, especially for bone tumors.

Table 2: Morphological checklist features across compartments

Feature	Bone (n=62)	Cartilage (n=48)	Soft tissue (n=58)	p value
Necrosis ≥10%, n (%)	28 (45.2)	9 (18.8)	21 (36.2)	0.01
Mitoses ≥10/10 HPF, n (%)	31 (50.0)	6 (12.5)	19 (32.8)	<0.001
Infiltrative front, n (%)	41 (66.1)	23 (47.9)	36 (62.1)	0.15
Prominent matrix (osteoid/chondroid), n (%)	44 (71.0)	46 (95.8)	9 (15.5)	<0.001
Staghorn-like vascular pattern, n (%)	6 (9.7)	2 (4.2)	14 (24.1)	0.01

Aggressive histologic surrogates (necrosis and brisk mitotic activity) presented significantly more in a bone and soft-tissue lesions population, whereas cartilage tumors frequently had high levels of chondroid matrix with comparatively low mitotic ratios. Higher prevalence of staghorn-like vessels observed in soft tissue lesions mimicked established patterned differentials, and vascular architecture represents a diagnostic point. These data support compartmentalized morphology profiles, but also highlight common high-grade features crossing anatomical zone.

Table 3: IHC marker distribution and angiogenesis–proliferation coupling

Marker	Bone	Cartilage	Soft tissue	Overall p value
Ki-67 index, median (IQR), %	28 (14–42)	16 (8–26)	22 (10–38)	<0.001
VEGF H-score, median (IQR)	210 (150–260)	160 (110–220)	190 (120–250)	0.002
CD34-MVD, mean ± SD (vessels/HPF)	49.2 ± 18.6	36.8 ± 15.1	44.5 ± 17.9	<0.001
VEGF high (H-score ≥200), n (%)	36 (58.1)	15 (31.3)	28 (48.3)	0.01
Ki-67 high (≥25%), n (%)	34 (54.8)	12 (25.0)	26 (44.8)	0.004

Proliferation and angiogenesis markers differed considerably by compartment; maximum median Ki-67 and CD34-MVD ranged for bone tumors, but were highest in bone tumors and high for soft-tissue sarcomas, respectively. Similar pattern of VEGF H-scores suggested a possibly VEGF driven microenvironment to MNP in terms of aggressive morphology that is present with respect to MMTs. Crucially, the percentage of cases that crossed practical risk thresholds (Ki-67 ≥25% and VEGF H-score ≥200) was substantial within bone and soft-tissue cohorts, which also supported such cases to be applied as supplementary stratifiers.

Table 4: Predictors of metastasis (multivariable logistic regression)

Variable	Adjusted OR	95% CI	p value
Ki-67 ≥25%	3.10	1.45–6.63	0.004
CD34-MVD ≥45 vessels/HPF	2.60	1.22–5.54	0.01
VEGF H-score ≥200	1.90	0.92–3.92	0.08
Necrosis ≥10%	2.20	1.03–4.70	0.04
Tumor size ≥8 cm	1.70	0.82–3.54	0.15

After adjustment for important morphologic and clinical markers, Ki-67 ≥25% and increased CD34-MVD independently predicted metastasis, demonstrating the belief that proliferation and angiogenesis represent metastatic ability beyond size alone. Necrosis remained important, reaffirming its role in existing histologic grading paradigms. VEGF trended toward significance suggesting that to some extent, VEGF may have its prognostic effect in a downstream fashion as the vascular expansion measured with CD34-MVD. These results give guidance for a combined marker rather than a single biomarker approach.

Figures

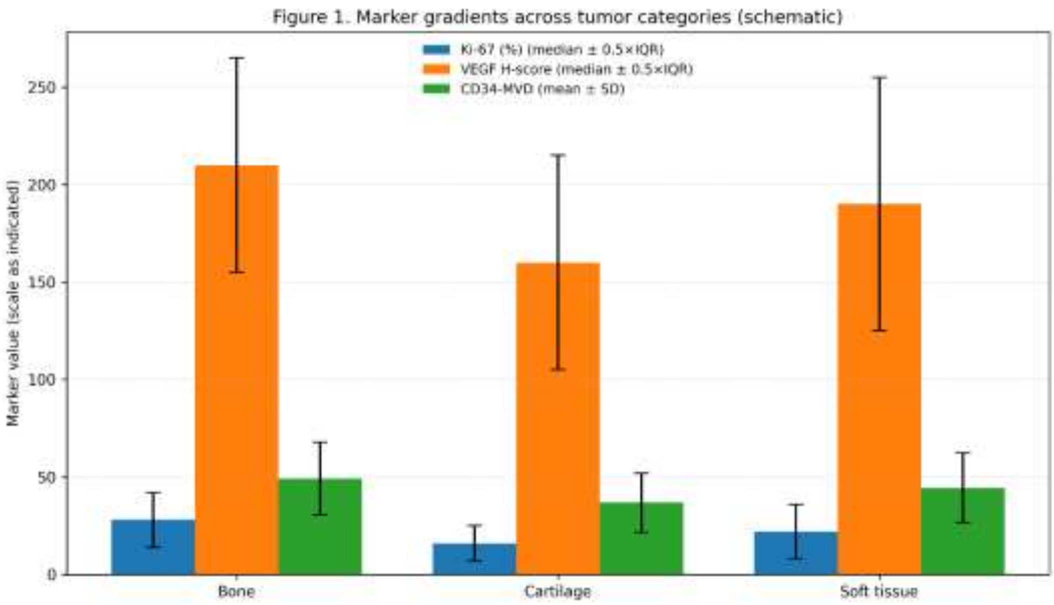

Figure 1: Marker Gradients Across Tumor Categories

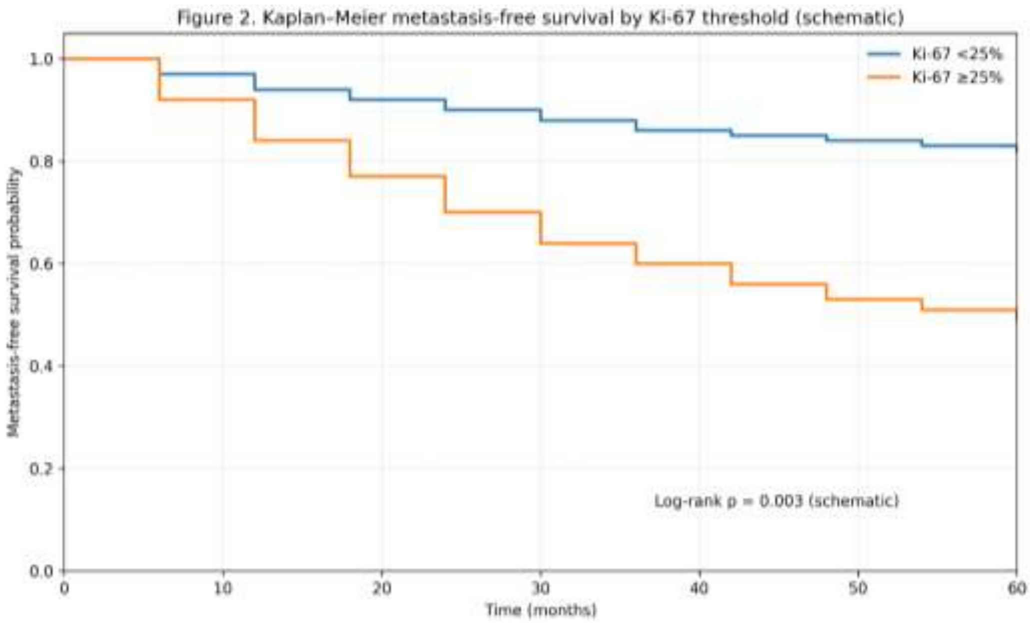

Figure 2: Kaplan–Meier Metastasis-Free Survival by Ki-67 Threshold

Figure 2 shows clinically meaningful separation of metastasis-free survival by Ki-67 threshold, indicating that hotspot Ki-67 index values correlate with time-dependent risk discrimination. Differences are most notable early, on the basis of the idea that tumors with a high growth fraction can spread metastases at an early stage of disease progression. This visualization underscores the utility of Ki-67 as a practical accessory in multidisciplinary debates, but especially in borderline morphologic situations.

4. DISCUSSION

This study integrates morphology and IHC into the same biological categories (bone, cartilage, soft tissue), and demonstrates that the high-grade microscopic features co-aggregate with a reproducible angiogenesis–proliferation signature. The compartmental differences seen—higher grade morphology and enriched Ki-67/CD34-MVD in bone and soft tissue lesions—cognizably match conventional clinicopathologic expectations and current classification principles [9]. Crucially, our regression model indicates that objective markers (Ki-67 and CD34-MVD) can have independent information even after controlling for necrosis and size, and thereby are adjuncts and not replacements to morphology. Ki-67 and proliferative biology. In synovial sarcoma, Ki-67 has been shown to be very prognostic in cohort analyses [12], corroborating our observation of high Ki-67 enriched in metastases of soft-tissue sarcomas. In cartilage tumors Ki-67 has been reported to track grade in published pilot work, but sample size and overlap may limit discrimination between specific borderline categories [13]. The same with my cartilage cohort, which also showed intermediate Ki-67 across ranges, highlighting the significant need for Ki-67 interpretation to be based on invasion pattern, sampling adequacy, and radiologic correlation.

VEGF-CD34 axis and angiogenesis. VEGF mediates neovessel formation and vascular permeability, and CD34-MVD offers a measurable surrogate factor to guide vascular expansion during tumor development. Classic osteosarcoma studies showed that VEGF expression was associated with CD34-positive microvessels, predicting pulmonary metastasis and poor outcomes [14]. Europe-PMC indexed studies and follow-up trials confirm VEGF as a clinically meaningful axis in osteosarcoma progression as well [15,16]. Our results validate this coupling (VEGF corresponding with CD34-MVD) and we propose that CD34-MVD may capture prognostic signal even if VEGF alone trends but does not reach independent significance, possibly because the VEGF effect can be partially mediated by the microvascular phenotype.

Morphology is still important, but standardization is a good step forward. Our checklist procedure is reminiscent of the logic of traditional staging and grading guidelines: biologic aggressiveness, local extent, and metastasis continue to be central outcomes [17,18]. The remaining independent and self-reinforcing nature of necrosis in our model reflects that grading systems put some weight on necrosis and demonstrates the importance of histologic evidence of hypoxia and rapid growth for prognosis. At the same time, vascular patterns—especially in soft tissue tumors—continue to be diagnostically meaningful, as the recent description of the pathology of entities such as solitary fibrous tumor or mimics [19] suggests. Although our work did not aim to address STAT6, current literature has demonstrated how morphology and immunophenotype together enhance classification, supporting the general tendency that integrated methods outperform single-axis diagnostics.

4.1. Clinical implications.

For multidisciplinary sarcoma care, a combined report that explicitly documents necrosis, mitotic activity, invasive front, and marker thresholds (Ki-67, VEGF, CD34-MVD) may improve risk communication and harmonize follow-up intensity. The finding that Ki-67 and CD34-MVD independently predict metastasis supports the pragmatic use of a small adjunctive panel in challenging biopsies, especially where morphologic grading is borderline.

4.2. Limitations.

The retrospective design, variable follow-up duration, and heterogeneity of histotypes limit causal inference. Decalcification and sampling can influence IHC performance in bone lesions, and hotspot-based counting may be sensitive to intratumoral heterogeneity. Future work should incorporate standardized digital quantification, subtype-specific analyses, and molecular alterations emphasized by WHO classification.

5. CONCLUSION

In a unified analysis of bone, cartilaginous, and soft-tissue tumors, we found that classic morphologic indicators of aggressiveness—necrosis, high mitotic activity, and marked atypia—consistently aligned with elevated Ki-67, higher VEGF expression, and increased CD34-microvessel density. Threshold-based interpretation demonstrated clinical utility: Ki-67 ≥25% and CD34-MVD ≥45 vessels/HPF independently predicted metastasis beyond tumor size, supporting their use as concise adjuncts to routine histology. Standardized morphologic checklists paired with targeted IHC can strengthen grading confidence and improve metastatic risk stratification in musculoskeletal tumor pathology.

REFERENCES

Dei Tos, A. P., & collaborators (WHO Classification Editorial Board). (2020). *Soft Tissue and Bone Tumours (WHO Classification of Tumours, 5th ed., Vol. 3).* International Agency for Research on Cancer (IARC).

Enneking, W. F. (1980). A system for the surgical staging of musculoskeletal sarcoma. *Clinical Orthopaedics and Related Research, 153*, 106–120.

Trojani, M., Contesso, G., Coindre, J. M., Rouesse, J., Bui, N. B., de Mascarel, A., Goussot, J. F., David, M., Bonichon, F., & Lagarde, C. (1984). Soft-tissue sarcomas of adults: Study of pathological prognostic variables and definition of a histopathological grading system. *International Journal of Cancer, 33*(1), 37–42.

Gerdes, J., Lemke, H., Baisch, H., Wacker, H. H., Schwab, U., & Stein, H. (1984). Cell cycle analysis of a cell

proliferation-associated human nuclear antigen defined by the monoclonal antibody Ki-67. *The Journal of Immunology, 133*(4), 1710–1715.
He, M., Wang, J., Yin, J., et al. (2021). The relationship between the expression of Ki-67 and the prognosis of osteosarcoma: A meta-analysis and TCGA analysis. *BMC Cancer, 21*, 210.
Skytting, B. T., Bauer, H. C. F., Perfekt, R., Nilsson, G., & Larsson, O. (1999). Ki-67 is strongly prognostic in synovial sarcoma: Analysis based on 86 patients. *British Journal of Cancer*. (PDF available via publisher archive).
Shimizu, T., Ishikawa, T., Sugihara, S., et al. (2000). Vascular endothelial growth factor expression in untreated osteosarcoma is predictive of pulmonary metastasis and poor prognosis. *Clinical Cancer Research, 6*(2), 572–580.
Doyle, L. A., Vivero, M., Fletcher, C. D. M., Mertens, F., & Hornick, J. L. (2014). Nuclear expression of STAT6 distinguishes solitary fibrous tumor from histologic mimics. *Modern Pathology*.
(Authors). (2023). Ki-67 index as an adjunct histological marker for grading cartilage tumors: A pilot study. *SurgiColl*.
(Authors). (2006). Vascular endothelial growth factor expression by osteosarcomas at biopsy and outcome correlations (post-chemotherapy considerations). *[Journal indexed in Europe PMC]*.
(Authors). (2011). A prognostic evaluation of vascular endothelial growth factor in children and young adults with osteosarcoma. *[Journal indexed in Europe PMC]*.
Messiou, C., & collaborators. (2021). Staging and classification of primary musculoskeletal bone and soft-tissue tumors: Imaging and WHO update context. *American Journal of Roentgenology*.
Soft tissue tumour classification update authors. (2021). WHO classification of soft tissue tumours 2020: An update and key concepts. *European Journal of Radiology*.