American Journal of
Biomedical Science & Research

www.biomedgrid.com
ISSN: 2642-1747

Research Article

Copyright © *Rana Naous*

Characterizing the Molecular Landscape of Fibrosarcoma: An Institutional Experience

Rana Naous*

Department of Pathology, University of Pittsburgh Medical Center, USA

***Corresponding author:** Rana Naous, Department of Pathology, University of Pittsburgh Medical Center, Pittsburgh, PA, USA.

To Cite This Article: *Rana Naous*. Characterizing the Molecular Landscape of Fibrosarcoma: An Institutional Experience. Am J Biomed Sci & Res. 2023 19(6) AJBSR.MS.ID.002655,* DOI: 10.34297/AJBSR.2023.19.002655

Received: August 18, 2023; **Published:** August 24, 2023

Abstract

Introduction: Fibrosarcoma is a very rare soft tissue sarcoma only diagnosed after excluding other mimicking entities with use of ancillary immunohistochemical and molecular testing.

Objective: Given their rarity, studies addressing the genetic landscape of fibrosarcoma are scarce. We analyzed our institutional fibrosarcoma cases in an attempt to further characterize the molecular landscape of fibrosarcoma and find potential therapeutic targets.

Methods: Targeted next-generation sequencing DNA analysis for characteristic DNA mutations and copy number variation was performed on 8 fibrosarcoma cases involving bone/soft tissue diagnosed at our institution between the years 2000 and present.

Results: Three out of our 8 cohort cases failed testing due to low DNA count. Among the remaining five cases, the most common genetic alteration was *CDKN2A/CDKN2B* copy number loss (4 cases, 80%), followed by TP53 mutations (3 cases, 60%). *NF1* mutation or copy number loss, *NOTCH* mutations and *BAP1* copy number loss had an equal frequency (2 cases each, 40%), and each of *SMAD4, TERT, FANCA, FAND2* and *PTCH1* alterations occurred at a frequency of 20%. *NOTCH, FANCA, FANCD2* and *PTCH1* mutations were reported as variants of uncertain significance (VUS).

Conclusion: Our study demonstrated that, similar to most sarcomas, the most common genetic aberrations in fibrosarcoma are *CDKN2A/CDKN2B* and *TP53* alterations. Additional novel alterations detected in our study with potential tumorigenesis role included *BAP1* and *NF1* copy number loss, *TERT* mutation, and *SMAD4* mutation. Additional large-scale studies are needed to further expand on the molecular landscape and biology of fibrosarcoma in search for potential implications on therapy.

Keywords: Fibrosarcoma, Molecular, Landscape

Introduction

Sarcomas are extremely rare tumors with less than 15,000 new cases diagnosed each year in the United States [1]. When compared to other tumor types, the predominant genomic aberrations in soft tissue and bone sarcomas are DNA copy number and chromosomal translocations; nonetheless, the number of targetable mutations remains considerably low [2,3]. Fibrosarcoma is a very rare soft tissue sarcoma that is only diagnosed after excluding other mimicking entities with the use of ancillary immunohistochemical and molecular testing [4]. Very few cytogenetic analyses have been reported on fibrosarcoma revealing multiple non-specific complex chromosomal anomalies, including balanced and unbalanced chromosomal translocations and copy number anomalies with no common or characteristic chromosomal changes to date [5-7]. Given their rarity, studies addressing the genetic landscape of fibrosarcoma are scarce. In our prior 2022 study [8] we demonstrated a gene fusion (*FNDC3B-PIK3CA*) of uncertain significance in 1 (10%) of 10 fibrosarcomas diagnosed at our institution in the past 20 years using next-generation sequencing (NGS) RNA fusion analysis while searching for novel gene fusion events, including NTRK-related fusions as described by *Yamazaki,*

This work is licensed under Creative Commons Attribution 4.0 License

et al., [9] who had reported the presence of novel neurotrophic receptor tyrosine kinase 3 (NTRK3) fusion in 2 of their fibrosarcoma cases. In the current study the author uses NGS DNA analysis on the corresponding institutional case cohort in search for characteristic DNA mutations and copy number variation in an attempt to further characterize the molecular landscape of fibrosarcoma and find potential therapeutic targets.

Materials and Methods

This study received Institutional Review Board exemption (MOD20080228-002). A detailed description of the case cohort used in this study has been provided in a prior publication [8]. The case cohort included eight pure fibrosarcoma cases involving bone or soft tissue diagnosed between the years 2000 and present with available formalin-fixed paraffin-embedded tissue for molecular testing. All cases demonstrated strict "fibrosarcoma" morphologic criteria as described by the WHO; a monomorphic fascicular spindle cell proliferation with no more than moderate degree of atypia and a non-specific immunophenotype [4]. Tumor grade was either low-grade (6 cases) or intermediate grade (2 cases). Ancillary immunohistochemical and molecular workup was performed to rule out morphologic mimickers. All cases were negative for cytokeratin, S100, desmin, EMA, CD34, and SMA (rare focal staining in 1 case). FISH for SYT gene rearrangement was negative in all cases. The above immunophenotype, along with the absence of SYT gene rearrangement, essentially excluded all morphologic mimics of fibrosarcoma including malignant peripheral nerve sheath tumor, solitary fibrous tumor, synovial sarcoma, biphenotypic sinonasal sarcoma, leiomyosarcoma, Kaposi sarcoma, and sarcomatoid carcinoma.

Targeted next-generation sequencing (NGS) DNA analysis was performed in accordance with standard clinical practice. Briefly DNA was extracted from manually-microdissected surgical FFPE tissue using the DNeasy Blood and Tissue Kit (Qiagen) and quantitated using the Glomax Discover fluorometer (Promega). DNA NGS libraries were generated using the DNA primers of the Oncomine Comprehensive Assay v3 (ThermoFisher Scientific) and quantitated using the TapeStation 4200 (Agilent Technologies) according to the vendor's protocol. NGS libraries were normalized and pooled for template preparation on the Ion Chef system (ThermoFisher Scientific) and subsequent sequencing on the Ion S5 GeneStudio instrument (ThermoFisher Scientific). Data analysis was performed using Torrent Suite v5.12 (ThermoFisher Scientific) and in-house software for DNA mutation, copy number variation, and tumor mutation burden calculation.

Results

The NGS DNA analysis results are summarized in Table 1. As previously mentioned, all 8 fibrosarcoma cases in our cohort demonstrated a similar morphology with monomorphic fascicular spindle cell proliferation, moderate atypia and variable mitosis (Figure 1A and B). The age range of our patients was 14 to 88 years old (median 60.5), with a male to female ratio of 1.6:1. Tumor location included head, jaw, upper and lower extremities, trunk and penis. Three out of our 8 cohort cases failed testing due to a low DNA count. Among the remaining five cases, the most common genetic alteration was *CDKN2A/CDKN2B* copy number loss (4 cases, 80%), followed by *TP53* mutations (3 cases, 60%). *NF1* mutation or copy number loss, *NOTCH* mutations and *BAP1* copy number loss had an equal frequency in our cohort (2 cases each, 40%). Each of *SMAD4*, *TERT*, *FANCA*, *FAND2* and *PTCH1* alterations occurred at a frequency of 20%. It is worth noting that some of these genetic aberrations may not represent defining events for tumorigenesis especially that some were reported as variants of uncertain significance (VUS), in particular *NOTCH*, *FANCA*, *FANCD2* and *PTCH1* mutations (Table 1).

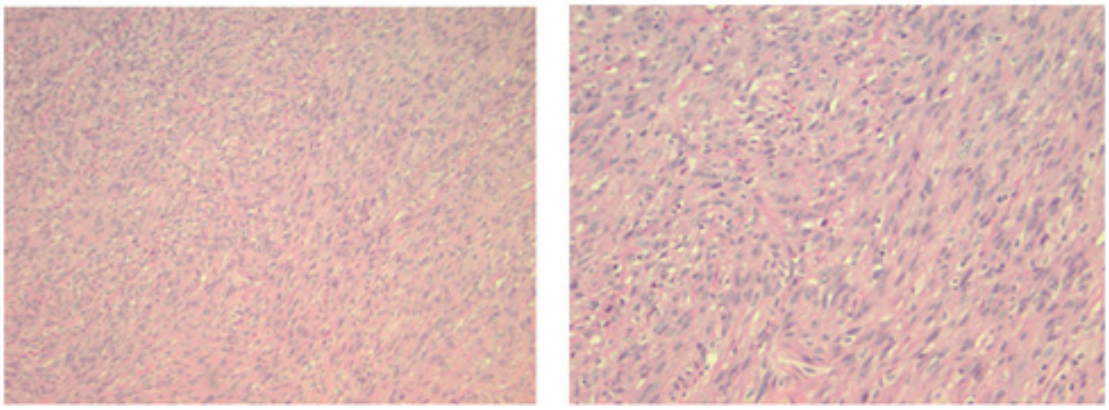

Figure 1(A,B): (A, B) H&E sections of case 5 demonstrating a relatively monomorphic fascicular spindle cell proliferation with moderate atypia set within a collagenous stroma (10x, 20x).

Table 1:

Case	Sex	Location	Age	Fibrosarcoma Grade	OCAv3 DNA Results
1	M	Right Thigh	68	Low-Grade	"CDKN2A, CDKN2B (chr9p) CN LOSS (CNR 0.17) TMB = 0 Mut/Mb"
2	F	Right Forearm	88	Intermediate-Grade	"TP53 c.743G>A p.R248Q 6.3% NOTCH1 c.368C>T p.T123M 49.6% (VUS) POLE c.6418delG p.E2140Rfs*62 54.5% (VUS) MAGOH c.346C>T p.P116S 6.5 % (VUS, lower coverage) NF1 c.1009G>A p.E337K 5.3 % (VUS, lower coverage) SETD2 c.5683G>A p.D1895N 5.3% (VUS, lower coverage) SMAD4 CN LOSS (CNR 0.39) chr8q (MYC, NBN) CN LOSS (CNR 0.51, 0.55) BAP1 CN LOSS (CNR 0.56) CDKN2A, CDKN2B (chr9p) CN LOSS (CNR 0.63, 0.58) NF1 CN LOSS (CNR 0.65)
3	F	Right Knee	67	Low-Grade	"NOTCH2 c.3625T>G p.F1209V 50.4% (VUS) FANCA c.953G>C p.R318T 35.4% (VUS) CDKN2A, CDKN2B (chr9p) CN LOSS (CNR 0.28, 0.31) NF1 CN LOSS (CNR 0.67) TMB = 3 Mut/Mb (26 percentile within all tumors)"
4	M	Penis	66	Low-Grade	"TERT c.1-124C>T p.C228T 37.9% (coverage, 29; 11 reads variant) TP53 c.524G>A p.R175H 46.4% FANCD2 c.28T>C p.S10P 36.3% (VUS) CDKN2A, CDKN2B (chr9p) CN LOSS (CNR 0.45, 0.52) TMB = 8 Mut/Mb (65 percentile within all tumors)"
5	M	Paraspinal	48	Intermediate-Grade	"TP53 c.839G>C p.R280T 63.7% PTCH1 c.1624C>T p.R542C 5.3% (VUS) BAP1 CN LOSS (CNR 0.30) TMB = 1 Mut/Mb (11 percentile within all tumors)"
6	M	Mandible	14	Low-Grade	FAILED, [DNA] too low (0.07ng/ul)
7	F	Right Abdominal wall	58	Low-Grade	FAILED, [DNA] too low (0.37ng/ul)
8	M	Left Middle Turbinate	43	Low-Grade	FAILED, [DNA] too low (0.04ng/ul)

Abbreviations: VUS: Variant of Uncertain Significance; CN: Copy Number; CNR: Copy Number Region; TMB: Tumor Mutation Burden

In our prior publication [8] case 2 was previously discussed and demonstrated a *FNDC3B-PIK3CA* gene fusion of uncertain significance, detected via NGS RNA fusion analysis. Of note, DNA/RNA sequencing performed by an outside institution was also reported on another case from our prior 2022 study cohort whereby BRAF p.G469A gene mutation was detected and mentioned in our prior manuscript with this mutation being most often reported in lung adenocarcinoma with an unknown prevalence in sarcoma. This case was removed from our current cohort.

Discussion

The most common genes altered in soft tissue sarcomas are *TP53* (47%), *CDKN2A* (22%), *RB1* (22%), *NF1* (11%), and *ATRX* (11%) with the majority of alterations in *TP53*, *NF1*, and *ATRX* being point mutations while the predominant alterations in *CDKN2A* and *RB1* being copy number losses [10]. In their study, *Bui, et al.,* tested all genomic alterations for their prognostic significance, and only *CDKN2A* alterations correlated significantly with prognosis. The study also showed that there was a trend to earlier recurrence with *CDKN2A* altered patients. In their analysis of 7733 soft tissue sarcoma patients, malignant peripheral nerve sheath tumors (MPNST), myxofibrosarcomas, and undifferentiated pleomorphic sarcomas showed a high prevalence of *CDKN2A* alterations.

CDKN2A loss has been shown previously to be a defining event for the malignant transformation of neurofibromas into MPNSTs [2,3]. One study [3] reported a prevalence of 26.3% of *CDKN2A* aberrations in fibrosarcoma; however, the authors do allude to the fact that their fibrosarcoma cohort had a limitation whereby pathology was not centrally reviewed, hence the fibrosarcoma diagnosis was solely made by the referring physician which may have led to incorrect or outdated diagnoses. *CDKN2A* (cyclin-dependent kinase inhibitor 2A) is a tumor suppressor gene that encodes two proteins: p16 and p14arf. The p16 protein is involved in cell cycle and senescence through the regulation of the cyclin-dependent kinase (CDK) 4/6 and cyclin D complexes, while p14arf activates *TP53*. Most common alterations are homozygous deletions, followed by inactivating mutations and promoter hypermethylation [11]. Further research is needed into the p16-CDK4-RB1 pathway and its role in targeted therapeutics.

CDKN2B serves as an important inhibitor of cell proliferation and cell cycle as it encodes for *CDKN2B* protein (p15, INK4B), which belongs to the INK4 class of cell cycle inhibitors [12]. *CDKN2B* lies adjacent to *CDKN2A* on human chromosome 9 and the entire CDKN2A-ARF-CDKN2B locus has shown to be frequently mutated or epigenetically silenced in many cancers [13-17]. In a study by *Scruggs, et al.,* [18], loss of *CDKN2B* was associated with an increase in myofibroblast differentiation and an increase in expression of myofibroblast-related transcription factors. The authors noticed that such effects were not observed with silencing *CDKN2A*. Although the actions of *CDKN2A* and *CDKN2B* are often considered redundant, being members of the INK4 family of cell cycle inhibitors with similar structural homology and binding affinity to CDK4/6, only *CDKN2B* appears to play a role in modulating fibroblast biology and fibroblast differentiation. The relationship of this unique *CDKN2B* role and its contribution in the biology or pathogenesis of fibrosarcoma remains to be elucidated.

TP53 is altered in 18.91% of soft tissue sarcoma patients [19]. This gene is mapped to chromosome 17 and plays a major role in regulating the response of mammalian cells to stresses and damage through the transcriptional activation of genes involved in cell cycle control, DNA repair, senescence, angiogenesis and apoptosis [20] Inactivation of the *p53* pathway may be derived from the *p53* mutation itself, or from other alterations, including increased expression of *MDM2* thus causing *p53* downregulation, or upstream secondary to *CDKN2A* mutation [21,22]. *P53* mutations are seen in most types of human cancer [23,24] and are one of the most prevalent genetic alterations in soft tissue sarcoma [25]. This is restated in our study, whereby the most common genetic alterations detected in our fibrosarcoma cohort were CDKN2A/CDKN2B and TP53 gene alterations.

BAP1 encodes a deubiquitinating hydrolase identified in 1998 that binds to the RING finger domain of the BRCA1 protein. *BAP1* is altered in 0.96% of malignant soft tissue neoplasms [19,26]. While not reported so far in fibrosarcoma, *BAP1* mutations are associated with many cancer types including uveal melanoma, mesothelioma, cutaneous melanoma, renal cell carcinoma, hepatocellular carcinoma, cholangiocarcinoma, meningioma, breast, thyroid, and lung cancer, among others [27].

NOTCH is altered in 2.1% of soft tissue sarcoma patients [19]. The Notch signaling pathway consists of four trans-membrane receptors, Notch1 to Notch4, that are encoded by homologous genes [28]. *NOTCH* signaling has been described in synovial sarcoma, Ewing sarcoma and rhabdomyosarcoma [29]. The relationship of *NOTCH* signaling in fibrosarcoma has not been described to date.

NF1 is altered in 5.66% of soft tissue sarcoma patients [19]. *NF1* mutations are identified in malignant peripheral nerve sheath tumor, myxofibrosarcoma, rhabdomyosarcoma, undifferentiated pleomorphic sarcoma and pleomorphic liposarcoma [30]. *RB1* is altered in 7.14% of fibrosarcoma patients [19]. RB1 gene encodes the cell cycle regulatory retinoblastoma gene protein (pRb), controls cellular differentiation during both embryogenesis and in adult tissues, regulates apoptotic cell death, maintains cell cycle arrest, and preserves chromosome stability. RB1 gene alterations are most commonly seen in leiomyosarcoma. It has been reported to a lesser extent in fibrosarcoma, rhabdomyosarcomas and pleomorphic sarcomas [31].

Telomerase reverse transcriptase (TERT) promoter gene mutation is identified in very few sarcoma cases including pleomorphic liposarcoma, dedifferentiated liposarcoma, myxoid liposarcomas, solitary fibrous tumor, undifferentiated pleomorphic sarcoma (UPS) and malignant granular cell tumor [19,32,33].

SMAD4 (*DPC4*) is a tumor suppressor gene located at chromosome 18q21.1 that belongs to the SMAD family, which mediates the TGF beta signaling pathway suppressing epithelial cell growth. Mutation of this gene is seen only rarely in soft tissue sarcomas [34]. Both *TERT* and *SMAD4* mutations have not been described in fibrosarcoma.

FANCA (Fanconi anemia, complementation group A) is a DNA repair gene, when mutated causes hypersensitivity to DNA damage with a greatly increased risk of multiple cancers [35]. *FANCA* mutation is the most frequent mutation in patients with Fanconi anemia but is less reported in patients with solid tumors [36]. *FANCA* is altered in 1.17% of soft tissue sarcoma patients [19]. *FANCD2* (Fanconi anemia, complementation group D2), similar to FANCA, is also involved in Fanconi anemia, a heterogeneous recessive disorder that causes cytogenetic instability, hypersensitivity to DNA crosslinking agents, increase in chromosomal breakage, and defective DNA repair. *FANCD2* is altered in 3.59% of soft tissue sarcoma patients [19]. Both FANCA and FANCD gene mutations have not yet been reported in fibrosarcoma.

PTCH1 is a tumor suppressor gene that is altered in 1.82% of sarcoma patients [19]. *PTCH1* alterations have been reported in intimal sarcoma, rhabdomyosarcoma, malignant peripheral nerve sheath tumor, angiosarcoma, extra skeletal osteosarcoma and extra skeletal myxoid chondrosarcoma [37]. Given that the *PTCH1* alteration in one of our fibrosarcoma case cohort was reported as a variant of uncertain significance, the relation between *PTCH1* aberrations and fibrosarcoma in general has yet to be investigated.

Conclusion

In conclusion, we attempted to characterize the molecular landscape of fibrosarcoma at our institution, and demonstrated that, similar to most sarcomas, the most common genetic aberrations in fibrosarcoma are *CDKN2A/CDKN2B* and *TP53* alterations. Additional novel alterations detected in our study with potential tumorigenesis role included *BAP1* and *NF1* copy number loss, *TERT* mutation, and *SMAD4* mutation. Overall, our study helps in further characterizing the mutational changes in fibrosarcoma with potential implications on therapy and prognosis. Additional large-scale studies are needed to further expand on the molecular biology of fibrosarcoma.

Declaration of Conflicting Interests

The author declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author received no financial support for the research, authorship and/or publication of this article.

Ethical Approval

This study was deemed to be exempt.

Informed Consent

Informed consent was not sought as the study spanned archived tissue from the past 20 years where the patients could not be reached.

Trial Registration

This study did not contain any clinical trials.

References

1. Siegel RL, Miller KD, Jemal A (2018) Cancer Statistics. CA Cancer J Clin 68: 7-30.
2. Nielsen GP, AO Stemmer Rachamimov, Y Ino, MB Moller, AE Rosenberg, et al. (1999) Malignant Transformation of Neurofibromas in Neurofibromatosis 1 Is Associated With Cdkn2a/P16 Inactivation. Am J Pathol 155(6): 1879-1884.
3. Sohier P, Armelle Luscan, Angharad Lloyd, Kevin Ashelford, Ingrid Laurendeau, et al. (2017) Confrmation of Mutation Landscape of Nf1-Associated Malignant Peripheral Nerve Sheath Tumors. Genes Chromosom Cancer 56(5): 421-426.
4. (2020) WHO Classification of Tumours Editorial Board. World Health Organization Classification of Soft Tissue and Bone Tumours, IARC Press 122-123.
5. Dal Cin P, Pauwels P, Sciot R, Van den Berghe H (1996) Multiple Chromosome Rearrangements in A Fibrosarcoma. Cancer Genet Cytogenet 87(2): 176-178.
6. Limon J, Szadowska A, Iliszko M, Babińska M, Mrózek K, et al. (1998) Recurrent Chromosome Changes in Two Adult Fibrosarcomas. Genes Chromosomes Cancer 21(2): 119-123.
7. Vanden Berg E, Molenaar WM, Hoekstra HJ, Kamps WA, de Jong B (1992) DNA Ploidy and Karyotype in Recurrent and Metastatic Soft Tissue Sarcomas. Mod Pathol 5(5): 505-514.
8. Leckey BD, John I, Wald A, Naous R (2022) Expanding the Molecular Genetic Spectrum of Bone and Soft Tissue Fibrosarcomas: An Institutional Experience. Int J Sur Pathol 30(2): 145-150.
9. Yamazaki F, Nakatani F, Asano N, Susumu Wakai, Masaya Sekimizu, et al. (2019) Novel NTRK3 Fusions in Fibrosarcomas of Adults. Am J Surg Pathol 43(4): 523-530.
10. Bui NQ, Przybyl J, Trabucco SE, Garrett Frampton, Trevor Hastie, et al. (2019) A Clinico-Genomic Analysis of Soft Tissue Sarcoma Patients Reveals CDKN2A Deletion as A Biomarker for Poor Prognosis. Clin Sarcoma Res 9: 12.
11. Collado M, Blasco MA, Serrano M (2007) Cellular Senescence In Cancer And Aging. Cell 130(2): 223-233.
12. Hannon GJ, Beach D (1994) p15INK4B is a Potential Effector of TGF-β-Induced Cell Cycle Arrest. Nature 371: 257-261.
13. Pavletich NP (1999) Mechanisms of Cyclin-Dependent Kinase Regulation: Structures of Cdks, their Cyclin Activators, and Cip and INK4 Inhibitors. J Mol Biol 287(5): 821-828.
14. Russo AA, Tong L, Lee JO, Jeffrey PD, Pavletich NP (1998) Structural Basis for Inhibition of The Cyclin-Dependent Kinase Cdk6 by the Tumour Suppressor p16INK4a. Nature 395: 237-243.
15. Drexler HG (1998) Review of Alterations of The Cyclin-Dependent Kinase Inhibitor INK4 Family Genes p15, p16, p18 and p19 in Human Leukemia-Lymphoma Cells. Leukemia 12(6): 845-859.
16. Ruas M, Peters G (1998) The p16INK4a/CDKN2A Tumor Suppressor and Its Relatives. Biochim Biophys Acta 1378(2): F115-F177.

17. Sharpless NE (2005) INK4a/ARF: A Multifunctional Tumor Suppressor Locus. Mutat Res 576(1-2): 22-38.

18. Scruggs AM, Koh HB, Tripathi P, Leeper NJ, White ES, et al. (2018) Loss of CDKN2B Promotes Fibrosis via Increased Fibroblast Differentiation Rather Than Proliferation. Am J Respir Cell Mol Biol 59(2): 200-214.

19. (2017) AACR Project GENIE: Powering Precision Medicine Through an International Consortium. Cancer Discov 7(8): 818-831.

20. Levine AJ (1997) p53, the Cellular Gatekeeper for Growth and Division. Cell 88(3): 323-331.

21. Bates S, Vousden KH (1999) Mechanisms of p53-Mediated Apoptosis. Cell Mol Life Sci 55(1): 28-37.

22. Liang Yin, Chun Xia Liu, Wei Xia Nong, Yun Zhao Chen, Yan Qi, et al. (2012) Mutational Analysis of p53 and PTEN in Soft Tissue Sarcoma. Mol Med Rep 5(2): 457-461.

23. Greenblatt MS, Hollstein M, Harris CC (1994) Mutations in the p53 Tumor Suppressor Gene: Clues to Cancer Etiology and Molecular Pathogenesis. Cancer Res 54(18): 4855-4878.

24. Hollstein M, Sidransky D, Vogelstein B, Harris CC (1991) p53 Mutations in Human Cancers. Science 253: 49-53.

25. Latres E, Drobnjak M, Pollack D, MR Oliva, M Ramos, et al. (1994) Chromosome 17 Abnormalities and TP53 Mutations In Adult Soft Tissue Sarcomas. Am J Pathol 145(2): 345-355.

26. Jensen DE, Proctor M, Marquis ST, HP Gardner, SI Ha, et al. (1998) BAP1: A Novel Ubiquitin Hydrolase Which Binds to The BRCA1 RING Finger and Enhances BRCA1-Mediated Cell Growth Suppression. Oncogene 16(9): 1097-1112.

27. Ewens KG, Lalonde E, Richards Yutz J, CL Shields, A Ganguly (2018) Comparison of Germline versus Somatic BAP1 Mutations for Risk of Metastasis in Uveal Melanoma. BMC Cancer 18(1): 1172.

28. Kopan R, Ilagan MX (2009) The Canonical Notch Signaling Pathway: Unfolding the Activation Mechanism. Cell 137(2): 216-233.

29. Rota R, Ciarapica R, Miele L, Locatelli F (2012) Notch Signaling In Pediatric Soft Tissue Sarcomas. BMC Med 10: 141.

30. Dodd RD, Mito JK, Eward WC, Chitalia R, Sachdeva M, et al. (2013) NF1 Deletion Generates Multiple Subtypes of Soft-Tissue Sarcoma That Respond to MEK Inhibition. Mol Cancer Ther 12(9): 1906-1917.

31. Kleinerman RA, Schonfeld SJ, Tucker MA (2012) Sarcomas In Hereditary Retinoblastoma. Clin Sarcoma Res 2(1): 15.

32. Campanella NC, Penna V, Abrahão Machado LF, Cruvinel Carloni A, Ribeiro G, et al. (2016) TERT Promoter Mutations in Soft Tissue Sarcomas. Int J Biol Markers 31(1): e62-e67.

33. Saito T, Akaike K, Kurisaki Arakawa A, Toda Ishii M, Mukaihara K, et al. (2016) TERT Promoter Mutations Are Rare in Bone and Soft Tissue Sarcomas of Japanese Patients. Mol Clin Oncol 4(1): 61-64.

34. Miyaki M, Kuroki T (2003) Role of Smad4 (DPC4) Inactivation in Human Cancer. Biochem Biophys Res Commun 306(4): 799-804.

35. Alter BP (2003) Cancer in Fanconi Anemia, 1927-2001. Cancer 97(2): 425-440.

36. Xia Q, Zhao LY, Yan YD, Liao Y, Di YS, et al. (2020) A Multiple Primary Malignancy Patient With FANCA Gene Mutation: A Case Report and Literature Review. Front Oncol 10: 1199.

37. Zehir A, Benayed R, Shah RH, Aijazuddin Syed, Sumit Middha, et al. (2017) Mutational Landscape of Metastatic Cancer Revealed from Prospective Clinical Sequencing of 10,000 Patients. Nat Med 23(6): 703-713.