

Manuscript Title

Immunotherapy as a Future Treatment for Relapsed and Metastatic Primary Pulmonary Sarcomas

Abstract

Primary pulmonary sarcomas (PPS) are aggressive tumors that are ideally treated with complete surgical resection, yet this approach is seldom possible due to disease state. Chemotherapy and radiotherapy have proven to be inadequate in the majority of cases. Due to the high rate of recurrence and advanced disease progression, novel approaches such as those presented by immunotherapy must be investigated to provide better prognosis. Studies predominantly focus on other soft tissue malignancies such as osteosarcoma, and research pertaining to PPS is limited.

Keywords: *pulmonary sarcoma, immunotherapy, relapse, metastasis*

Primary pulmonary sarcomas (PPS) are exceedingly uncommon,^{1,2} but the increase in radiation exposure as a risk factor due to cancer treatment and otherwise leaves an increase in incidence to speculation.² Arising from the lung parenchyma, bronchi and pulmonary vasculature,³ the three most common types include leiomyosarcoma, malignant fibrous histiocytoma and synovial sarcoma.^{4,5}

PPS often remains asymptomatic, but may present with the same symptoms as lung carcinoma^{4,5} such as atelectasis, cough with expectoration and hemoptysis.⁶ Diagnosis occurs when a sarcoma of an extrathoracic location is ruled out, as metastatic sarcoma to the lungs is much more common.⁷ Computed tomography imaging is crucial as a diagnostic modality in identifying these thoracic lesions,^{1,8} which may include areas of stippled calcification and focal areas of necrosis.^{3,9} Immunohistochemistry for each sarcoma differs and overall structure is important in determination of grade and stage.³ The ideal management of disease is through curative surgical resection,⁷ yet is seldom possible.⁸ Radiation and chemotherapy are used as therapy in inoperable cases, relapse and metastatic disease.^{2,7} However, these are not nearly as effective,^{2,3} with a median survival of only 10-15 months.¹⁰ In synovial sarcoma, the translocation of t(X;18)(p11.2;q11.2) results in the fusion products of either SYT-SSX1 or SYT-SSX2, in which SYT-SSX1 variant is associated with a higher mitotic rate and worse prognosis.² Prognostic factors such as this have been associated with increasing rates of successive relapse.³ As this disease group is aggressive, and optimal therapies are either not possible or inadequate, novel approaches are necessary to improve prognosis.

Such a therapy in recent focus is immunotherapy. The basis for this approach stems from the observation made by Coley in 1891, when a small cell sarcoma recurrence receded following erysipelas infection and further local injections of pathogen.^{11,12} This was thought to occur as the malignant cells were caught in crossfire between the immune system and pathogen, subject to the antitumor effects elicited in the process.¹¹ This is supported with the fact that sarcomas occur three-fold more often in immunosuppressed patients when compared to the general population^{13,14} as well as the stability of present disease with the use of non-specific immunomodulators.^{15,16}

High-dose treatment with interleukin-2 (IL-2) had been investigated in numerous studies.¹⁵ Stimulation of T and natural killer (NK) cells occurs, which are essential in the immune control of malignancy. This modality of treatment has been FDA-approved for the treatment of metastatic melanoma and renal cell carcinoma, and had been used in therapy for sarcoma in a limited number of studies.¹⁶ Studies in the relatively distant past had demonstrated a response rate ranging from 0% to 33%, but limited by very small sample sizes.^{17,18} More recent studies concerning metastatic solid tumors of various etiologies treated with IL-2 had demonstrated an unlikely relapse after a follow-up period of 30 months without disease.¹⁶ This indicates that IL-2 has potential in the arsenal of therapy for recurrent or metastatic PPS that should be revisited with updated delivery techniques and manipulation.

Interferon had been investigated in the past as a potential immunotherapy for malignancy¹⁵ by potentiating immune cells and increasing presentation of antigens to T lymphocytes.¹⁵ This antitumor activity is demonstrated by a report of cases in which more than half of patients with osteosarcoma receiving interferon therapy had partial responses.¹⁹ Similarly, a study in 1989 by Edmonson *et al*²⁰ demonstrated a 15% response rate using interferon for patients with metastatic bone sarcomas. In 2003 Roozendaal and colleagues²¹ described a case of primary sarcoma in the calf with pulmonary metastasis refractory to treatment with chemotherapy. High-dose chemotherapy followed by

peripheral stem cell transplantation was considered too toxic, thus the patient was initiated on interferon therapy. After six months the patient showed a significant partial response. However, these interferon studies predominantly regard osteosarcoma,¹⁶ and more studies pertaining to the sarcoma subtypes of pulmonary origin are needed.

Another immunostimulatory therapy is liposomal-muramyl tripeptide phosphatidyl-ethanolamine (L-MTP), which is a synthetic analog of the bacterial cell wall.¹⁵ Studies have shown that L-MTP increases tumor cell killing *in vitro* via mononuclear cells when compared to control groups, and was quantified with the measurement of TNF alpha IL-6 as immunomodulatory markers.¹⁶ Furthermore, L-MTP has been shown in laboratory models to reduce the metastatic tumor burden, demonstrating a possible approach in aggressive and advanced malignancies as discussed.²² Unfortunately, this approach has predominantly been investigated with focus on osteosarcoma, yet has potential for other soft tissue malignancies such as PPS.²³ L-MTP may similarly to IL-2 and interferon function as a non-specific immunomodulator in cancer treatment.

An interesting therapy in development is the vaccination approach. Vaccines targeting gangliosides GD2, GD3 and GM2 in patients with solitary metastases excised in combination with GM-CSF or IL-2 have the potential to stimulate the immune response to lysate sarcoma cells.^{16,24} Furthermore, observations have exhibited that this therapy has no major toxicities,²⁴ an important advantage over chemo-radiation. A phase II trial using bivalent vaccination with GD2 and GD3 has been conducted demonstrating a positive serologic response when compared to control groups in patients with surgically disease-free, receiving adjuvant therapy and are still being followed-up for survival and presence of recurrence (NCT01141491).^{16,25,26} Although studies focus on neuroblastoma and other sarcomas, vaccination is of great interest for PPS as they express a high extent of GD2 and GD3 expression.¹⁵ More specifically targeted to synovial sarcomas, a vaccine has been developed using HLA-A24-optimized SYT-SSX peptide.^{16,27} Bloom *et al*²⁷ had completed a trial determining that this vaccine could be safely administered to synovial sarcoma patients, resulting in a significantly stabilizing aggressive disease. Stability was documented with a two-fold increase in cytotoxic T lymphocytes in the group receiving the vaccination and lack of progression in serial computed tomography scans.²⁷ Vaccination seems to be a developing future direction of treatment for aggressive sarcomatous disease.

Cancer testis antigens are proteins that are not expressed on normal tissue and are typically found in association with sarcoma tumor cells and germline tissues.¹⁵ These antigens are recognized by cytotoxic T lymphocytes¹⁵ and are thus possible targets when treating sarcomas. More specifically, 80% of synovial sarcomas express NY-ESO,^{15,28} 100% of synovial sarcomas express PRAME²⁹ and 14% of non-uterine leiomyosarcomas express MAGE-A3.³⁰ Such an approach would involve introducing adoptive transfer of autologous T cells expressing the aforementioned peptides, which can reach a response rate of 50% with persistent engineered T cell persistence.³¹

Evidently, immunotherapy is a direction that holds much promise for the treatment of relapsing or metastatic sarcomas, including that of primary pulmonary sarcoma. Investigation as to this modality of intervention is necessary as a rise in pulmonary sarcomas may have a delayed association with the use of radiotherapy for other thoracic and chest wall malignancies.

References

1. Aribas OK, Gormus N. (2001) Obstructing endobronchial malignant fibrous histiocytoma. *European Journal of Cardio-thoracic Surgery* 19:716-718
2. Etienne-Mastroianni B, Falchero L, Chalabreysse L, *et al.* (2002) Primary sarcomas of the lung. A clinicopathologic study of 12 cases. *Lung Cancer* 38:283-289
3. Weiss textbook
4. Kasturi S, Rendla SB, Vemula S, Raju P S. (2014) Primary pulmonary synovial sarcoma: a rare case report. *Int J Res Med Sci* 2(4):1729-1731
5. Keel SB, Bacha E, Mark EJ, Nielsen GP, Rosenberg AE. (1999) Primary pulmonary sarcoma: a clinicopathologic study of 26 cases. *Mod Pathol* 12(12):1124-1131
6. Nascimento AG, Unni KK, Bernatz PE. (1982) Sarcomas of the lung. *Mayo Clinic Proc* 57:335-359
7. Goncalves MJ, Mendes MM, Joao F, Lopes JM, Honavar M. (2011) Primary pleomorphic sarcoma of lung – 11 year survival. *Rev Port Pneumol* 17(1):44-47
8. Falkenstein-Ge RF, Kimmich M, Grabner A, Horn A, Friedel G, Ott G, Kohlhauf M. (2014) Primary pulmonary synovial sarcoma: a rare primary pulmonary tumor. *Lung* 192(1):211-214
9. Shirai T, Tsuchida S, Terauchi R, Mizoshiri N, Konishi E, Tomita Y, Shimada J, Fujiwara H, Kubo T. (2014) Primary pulmonary synovial sarcoma requiring differentiation from pulmonary metastasis of tibial adamantinoma: a case report. *BMC Res Notes* 7:736

10. Van Glabbeke M, Van Oosterom AT, Oosterhuis JW, *et al.* (1999) Prognostic factors for the outcome of chemotherapy in advanced soft tissue sarcoma: an analysis of 2,185 patients treated with anthracycline- containing first-line regimens—a European organization for research and treatment of cancer soft tissue and bone sarcoma group study. *Journal of Clinical Oncology* 17(1):150-157
11. Coley WB. (1891) II. Contribution to the knowledge of sarcoma. *Annals of Surgery* 14:199-220
12. Wiemann B, Starnes CO. (1994) Coley's toxins, tumor necrosis factor and cancer research: a historical perspective. *Pharmacology and Therapeutics* 64(3):529-564
13. Gatti RA, Good RA. (1971) Occurrence of malignancy in immunodeficiency disease. A literature review. *Cancer* 28(1):89-98
14. Penn I. (1995) Sarcomas in organ allograft recipients. *Transplantation* 60(12):1485-1491
15. D'Angelo SPD, Tap WD, Schwartz GK, Carvajal RD. (2014) Sarcoma Immunotherapy: Past Approaches and Future Directions. *Sarcoma* Article ID 391967, 13 pages. <http://dx.doi.org/10.1155/2014/391967>
16. Pollack SM, Logers ET, Rodler ET, Yee C, Jones RL. (2011) Immune-Based Therapies for Sarcoma. *Sarcoma* Article ID 438940, 7 pages. doi:10.1155/2011/438940
17. Rosenberg SA, Lotze MT, Yang JC, *et al.* (1989) Experience with the use of high-dose interleukin-2 in the treatment of 652 cancer patients. *Annals of Surgery* 210(4):474-485
18. Schwinger W, Klass V, Benesch M, *et al.* (2005) Feasibility of high-dose interleukin-2 in heavily pretreated pediatric cancer patients. *Annals of Oncology* 16(7):1199-1206
19. Ito H, Murakami K, Yanagawa T. (1980) Effect of human leukocyte interferon on the metastatic lung tumor of osteosarcoma: case reports. *Cancer* 46(7):1562-1565
20. Edmonson JH, Long HJ, Frytak S, Smithson WA, Itri LM. (1987) Phase II study of recombinant alfa-2a interferon in patients with advanced bone sarcomas. *Cancer Treatment Reports* 71(7-8):747-748
21. Roozendaal KJ, de Valk B, ten Velden TAA, van der Woude HJ, Kroon BBR. (2003) Alveolar soft-part sarcoma responding to interferon alpha-2b. *British Journal of Cancer* 89:243-245
22. Zhu L, McManus MM, Hughes DPM. (2013) Understanding the biology of bone sarcoma from early initiating events through late events in metastasis and disease progression. *Front Oncol* 3:230
23. Uehara T, Fujiwara T, Takeda K, Kunisada T, Ozaki T, Udono H. (2015) Immunotherapy for Bone and Soft Tissue Sarcomas. *BioMed Research International* Article ID 820813, 11 pages. <http://dx.doi.org/10.1155/2015/820813>
24. Kushner BH, Cheung IY, Modak S, Kramer K, Ragupathi G, Cheung NV. (2014) Phase I Trial of Bivalent Gangliosides Vaccine in Combination with •-Glucan for High-Risk Neuroblastoma in Second or Later Remission. *Clin Cancer Res* 20(5):1375-1382
25. Carvajal RD, Agulnik M, Ryan CW, Milhem MM, George S, *et al.* (2014) Trivalent ganglioside vaccine and immunologic adjuvant versus adjuvant alone in metastatic sarcoma patients rendered disease-free by surgery: A randomized phase 2 trial. *J Clin Oncol* 32:5s, (suppl; abstr 10520)
26. Slovin SF, Keding SJ, Ragupathi G. (2005) Special Feature: Glycobiology Of Xenotransplantation And Cancer Part I. *Immunology and Cell Biology* 83:418-428
27. Bloom JE, McNeel DG, Olson BM. (2012) Vaccination using peptides spanning the SYT-SSX tumor-specific translocation. *Expert Rev Vaccines* 11(12):1401-1404
28. Jungbluth AA, Chen YT, Stockert F, *et al.* (2001) Immunohistochemical analysis of NY-ESO-1 antigen expression in normal and malignant human tissues. *International Journal of Cancer* 92(6):856-860
29. Skubitz KM, Pambuccian S, Carlos JC, Skubitz APN. (2006) Identification of heterogeneity among soft tissue sarcomas by gene expression profiles from different tumors. *Journal of Translational Medicine* 6:article 23
30. Ayyoub M, Taub RN, Keohan M-L, *et al.* (2004) The frequent expression of cancer/testis antigens provides opportunities for immunotherapeutic targeting of sarcoma. *Cancer Immunity* (4):article 7
31. D'Angelo S, Merchant M, Melchiori L, Zhang H, Pandite L, Holdich T, Binder-Scholl G, *et al.* (2015) Optimizing engineered TCR T cell therapy for synovial sarcoma. *Journal of ImmunoTherapy of Cancer* 3:P159