Chapter

Current Diagnostic Techniques in Sarcoidosis

Rajarajan Anandavelu and Ahmed Fahim

Abstract

Sarcoidosis is an enigmatic disorder with a propensity for lung involvement in the majority of cases (~90%) and is characterized by noncaseating granulomatous inflammation on histological analysis. The techniques to establish the diagnosis have evolved over time, and a clear diagnostic algorithm for the clinicians dealing with this disease is desirable. Thoracic computed tomography is the imaging modality of choice in pulmonary sarcoidosis and provides accurate assessment of staging, parenchymal involvement, and response to immunomodulatory therapies. The advent of EBUS-TBNA has been a step forward with an excellent diagnostic yield in the presence of mediastinal/hilar lymphadenopathy and has replaced the traditional approach of obtaining biopsy samples via transbronchial and endobronchial routes. The preferred initial investigation for the confirmation of diagnosis is dependent on the organ involvement and the expertise available. A core biopsy of cervical lymph nodes is a less invasive and economical alternative in selected cases of suspected pulmonary sarcoidosis and warrants further evaluation in prospective manner to establish if it can be considered as a first-line investigation in all new cases suspected to have pulmonary sarcoidosis. A multidisciplinary approach is crucial for the diagnosis and management, and a simplified algorithm is proposed to help guide clinicians dealing with this disease of myriad clinical and radiological manifestations.

Keywords: sarcoidosis, granulomatous inflammation, lymphadenopathy, core biopsy, endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA)

1. Introduction

Sarcoidosis is an enigmatic disorder with a propensity for lung involvement in the majority of cases (90%) and is characterized by noncaseating granulomatous inflammation on histological analysis. The techniques to establish the diagnosis have evolved over time, and a clear diagnostic algorithm for clinicians dealing with this disease is desirable. Thoracic computed tomography is the imaging modality of choice in pulmonary sarcoidosis and provides accurate assessment of the stage, parenchymal involvement, and response to immunomodulatory therapies. The advent of EBUS-TBNA has been a step forward with an excellent diagnostic yield in the presence of mediastinal/hilar lymphadenopathy and has replaced the traditional approach of obtaining biopsy samples via transbronchial and endobronchial routes. The preferred initial investigation for the confirmation of diagnosis is dependent upon the organ involved and the expertise available. A core biopsy of cervical lymph nodes is a less invasive and economical alternative in selected cases

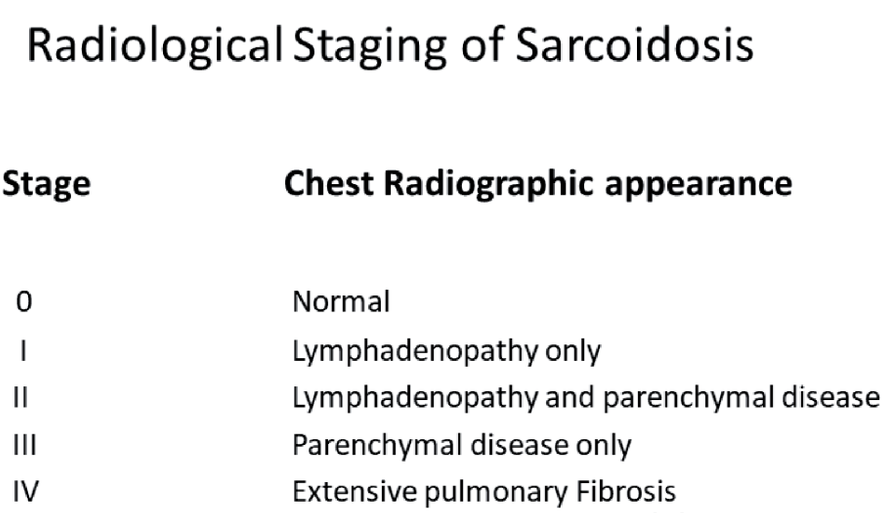

Figure 1.
Chest radiograph appearances according to stage of sarcoidosis.

of suspected pulmonary sarcoidosis and warrants further evaluation in a prospective manner to establish if it can be considered a first-line investigation in all new cases suspected to have pulmonary sarcoidosis. A multidisciplinary approach is crucial for the diagnosis and management; a simplified algorithm is proposed to help guide clinicians dealing with this disease of myriad clinical and radiological manifestations.

Sarcoidosis is a disease of uncertain etiology characterized by evidence of non-necrotizing granulomatous inflammation on histological assessment. As the disease commonly affects the lungs, the patients are likely to be referred to pulmonologists for further investigations. Chest radiography is usually the first investigation carried out in the primary care setting suggesting the possibility of hilar lymph node enlargement, and sarcoidosis has traditionally been staged according to chest radiographic appearances (**Figure 1**).

Radiological assessment of sarcoidosis has been revolutionized by high-resolution computed tomography (HRCT) scanning of lungs and is currently the best available modality to assess the extent of involvement of lung parenchyma/interstitial compartment and response to immunomodulatory therapies to evaluate alveolitis and reversibility of the disease process. The current diagnostic techniques for histological assessment in pulmonary sarcoidosis are invasive, and there is a need for a less invasive approach to obtain tissue sample(s). This chapter aims to discuss the available diagnostic techniques in sarcoidosis and propose an algorithm to the pulmonologists/radiologists and clinicians dealing with sarcoidosis as a tool to aid for reaching the diagnosis with minimally invasive investigations. It is recommended that an ultrasound-guided core biopsy of cervical lymph nodes (along with assessment of parotid glands) may be considered first-line investigation in the presence of mediastinal and or hilar lymphadenopathy, a suitable target to sample neck lymph nodes.

2. Diagnostic techniques

The diagnosis of sarcoidosis is best supported by the presence of noncaseating/necrotizing granulomatous inflammation on histological analysis, following the exclusion of other granulomatous disorders such as mycobacterial or fungal diseases on special immunohistochemical stains. The diagnostic techniques for sarcoidosis can be subclassified into four groups:

- Radiological techniques
- Bronchoscopic techniques
- Ultrasound-guided biopsy techniques
- Surgical techniques

2.1 Radiological imaging techniques

These include chest radiography, high-resolution computed tomography, and positron emission tomography (PET) scanning.

2.1.1 Chest radiography

The typical chest radiographic appearances in sarcoidosis consist of symmetric bilateral hilar and mediastinal lymphadenopathy. Fibrosis is noticed in 5–25% of patients with sarcoidosis on initial chest radiograph [1]. **Figure 2** demonstrates an example of advanced stage sarcoidosis with the development of fibrosis and parenchymal distortion.

2.1.2 High-resolution computed tomography

High-resolution computed tomography has improved the diagnostic accuracy of sarcoidosis in terms of parenchymal involvement (**Figure 3**) and assessed any reversible component such as alveolitis that may not be readily evident on chest radiography. Moreover, the abnormalities on HRCT scan do correlate better with respiratory functional impairment than chest radiograph findings [2].

Sarcoidosis is a disease with a myriad of radiological abnormalities on HRCT, including hilar and mediastinal lymphadenopathy, ground glass abnormality, and fibrosis in a peribronchovascular, perilymphatic distribution, pulmonary parenchymal nodules, and beading of the fissures. There is a mid- to upper-zone preponderance of these abnormalities and usually distributed along the bronchovascular

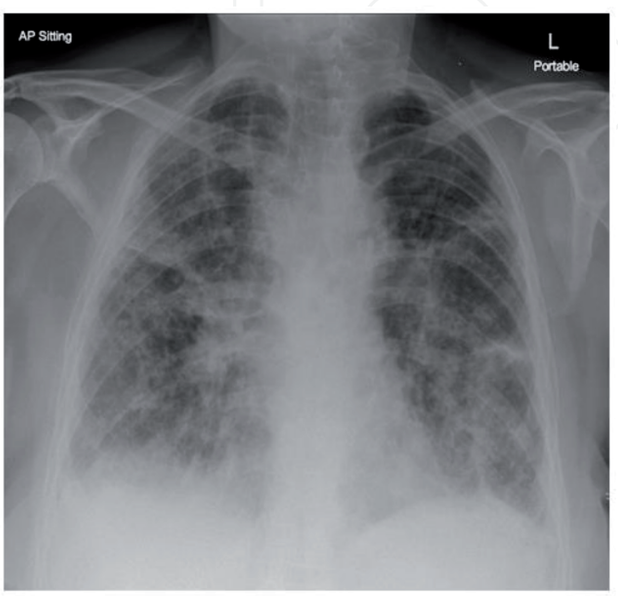

Figure 2.
Stage III sarcoidosis with evidence of hilar lymphadenopathy and parenchymal involvement.

bundles [3]. HRCT may be able to demonstrate lymphadenopathy in mediastinal distribution better than a chest radiograph and sometimes demonstrate evidence of calcification within these nodes (**Figure 4**).

The precise role of HRCT in the clinical monitoring of sarcoidosis is unknown. However, it may prove to be a useful tool to assess acute alveolitis and inflammation in selected cases of refractory sarcoidosis, where treatment decisions to commence

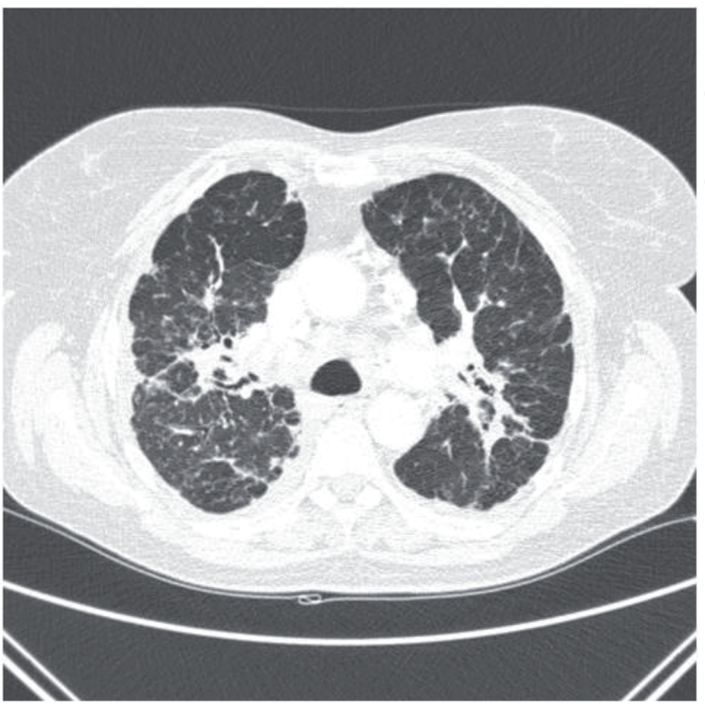

Figure 3.
Parenchymal distortion with fibrosis in a peribronchovascular distribution in bilateral upper lobes in sarcoidosis.

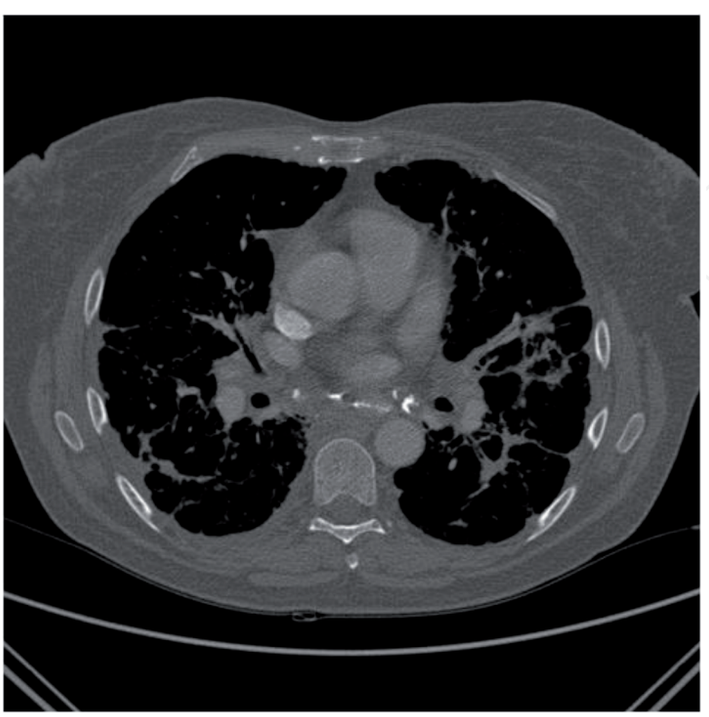

Figure 4.
Calcified mediastinal lymphadenopathy with bilateral hilar nodal enlargement in a patient with confirmed sarcoidosis.

biologic therapies such as infliximab are being made [3]. A study by De Boer et al. showed that the total extent of parenchymal disease on the CT scan on a lobar basis could predict the likelihood of transbronchial biopsy being positive following bronchoscopy [4], demonstrating its utility on diagnostic grounds.

2.1.3 Positron emission tomography scan

Positron emission tomography scan can be a useful tool to detect the extent of the disease, identify multisystem disease such as cardiac sarcoidosis, and may help to identify a desirable site for biopsy [5]. Moreover, it could be invaluable in the decision to initiate immunosuppression and assess the efficacy of treatment [6, 7]. Furthermore, it may help in predicting relapse in pulmonary sarcoidosis [8].

A retrospective study by Teirstein et al. showed that a combination of diagnostic modalities such as 18F-fluorodeoxyglucose (FDG-PET) and CT scan is more sensitive than PET-only imaging [9]. Whole-body FDG-PET was found to be significantly better in identifying occult and reversible granulomas. Moreover, a positive PET scan in isolation should not be considered as an indication for treatment. In another study by Yu et al., the sensitivity and specificity for benign and malignant disease were 94.2% and 73.8%, respectively [10]. It was, however, noted that maximum standard uptake value (SUVMax) as semiquantitative measurement alone could not be used to differentiate benign vs. malignant lesions.

The FDG-PET scan has a cumulative effect in cardiac sarcoidosis. PET scan has also been evaluated in predicting supraventricular arrhythmias, and it was noted that patients with left atrial enlargement were associated with increased likelihood of supraventricular arrhythmias [11]. Smedema et al. reported that biventricular late gadolinium enhancement was the strongest predictor of adverse outcome, and an asymptomatic myocardial scar of less than 8% in the left ventricular mass was associated with a favorable outcome in patients with pulmonary sarcoidosis [12].

On the basis of current available evidence, the role of PET-CT is limited in routine clinical care of patients with pulmonary sarcoidosis. However, it may be a useful imaging modality in multisystem sarcoidosis, in particular when the clinical suspicion for cardiac involvement is high and the diagnostic techniques such as echocardiography and or cardiac MRI have unequivocal results. Moreover, PET-CT may become a useful adjunct to assess the response to immunosuppression with corticosteroids and/or antimetabolites and may guide us to an appropriate biopsy site to sample suspected multisystem disease.

2.2 Bronchoscopic techniques

2.2.1 Bronchoalveolar lavage (BAL) and transbronchial biopsy (TBB)

Bronchoscopic techniques have been employed in the evaluation of pulmonary sarcoidosis for a very long time and have been the mainstay of histological confirmation historically. Granulomatous inflammation in sarcoidosis usually involves the bronchovascular and centrilobular structures.

Transbronchial biopsies help to obtain the histological diagnosis in support of clinical-radiological diagnosis especially when the superficial mucosal or cutaneous lesions are not amenable for sampling [13, 14]. The diagnostic sensitivity of TBB in the diagnosis of a broad spectrum of interstitial lung diseases (ILDs) ranges from 29 to 79% [15–19]. The British Thoracic Society Sarcoidosis Registry data has previously showed that transbronchial biopsies have lesser diagnostic yield than EBUS-TBNA [20].

Bronchoalveolar lavage findings supportive of sarcoidosis include predominant lymphocytosis on differential cell count analysis along with CD4/CD8 lymphocyte ratio of more than 1. Müller-Quernheim et al. demonstrated that inflammation is compartmentalized in sarcoidosis resulting in lymphocyte abundance in the involved organs [21]. In a study by Prasse et al., patients with sarcoidosis had higher expression of IL2, IFN gamma, and TNF alpha [22]. Furthermore, Tanriverdi et al. showed that high CD4/CD8 ratio, though specific is not a sensitive test for the diagnosis of sarcoidosis, and therefore, it does require clinico-radiological and pathological correlation [23]. BAL lymphocytosis of ≥40% in the appropriate clinical context would support the diagnosis of hypersensitivity pneumonitis or cellular nonspecific interstitial pneumonia (NSIP) over sarcoidosis [24]. However, bronchoalveolar lavage findings in isolation are unlikely to help establish the diagnosis of sarcoidosis.

2.2.2 Endobronchial ultrasound-guided transbronchial needle aspiration (EBUS-TBNA)

EBUS-TBNA has been significant in the diagnostic pathway of sarcoidosis and has obviated the need for a TBB (in most cases where mediastinal adenopathy is present) with associated risk of pneumothorax. EBUS-TBNA is a safe minimally invasive option and is the preferred diagnostic procedure before surgical techniques such as mediastinoscopy are considered for sampling mediastinal nodes [25].

Kitamura et al. demonstrated a sensitivity of 87.5% for combined cytological and histological examination [26]. A prospective study by Oki et al. compared the diagnostic yield of EBUS-TBNA and TBLB through a flexible bronchoscope in patients with stage I and II of sarcoidosis [27]. The diagnostic yield was 94% (stage I, 97%; stage II, 88%) and 37% (stage I, 31%; stage II, 50%), respectively; the complications such as pneumothorax and moderate bleeding were noted in patients, who underwent TBLB, albeit one case of pneumothorax and three cases of moderate bleeding among a total of 62 patients were seen.

A randomized controlled trial evaluated the use of endosonographic nodal aspiration against bronchoscopic biopsy, among patients with suspected stage I/II pulmonary sarcoidosis [28]. The diagnostic yield to detect granulomas for endosonography was 80% (95% CI, 73–86%), in comparison to 53% (95% CI, 45–61%) for bronchoscopy cohort ($P < 0.001$), suggesting a significantly higher diagnostic yield with endosonographic procedures. On the other hand, a randomized controlled trial by Gupta et al. showed that the diagnostic yield in sarcoidosis by conventional TBNA along with endobronchial biopsy (EBB) and TBLB is similar to EBUS-TBNA with TBLB [29].

The diagnostic accuracy of EBUS-TBNA with rapid on-site evaluation (ROSE) was compared to the final cytological assessment and to TBLB and EBB in a prospective study, and it showed that sensitivity for EBUS-TBNA with ROSE was 87.8% (specificity 91%, positive predictive value 97.7%) and concluded that it should be considered as first-line investigation for the evaluation of mediastinal adenopathy [30].

In patients with predominant mediastinal and/or intra-abdominal lymph nodes, which is not amenable for EBUS procedure, endoscopic ultrasound-guided fine aspiration (EUS-FNA) can be a potential option; Michael et al. demonstrated in a retrospective study that [31] EUS-FNA was able to diagnose sarcoidosis in 86% of cases (n = 18 of 21) and was able to rule out recurrence of malignancy in 75% (three out of four cases).

2.2.3 Transbronchial lung cryobiopsy (TBLC)

Transbronchial lung cryobiopsy is currently being increasingly considered as a diagnostic tool in ILD. The procedure requires general anesthesia and fluoroscopic

guidance for sampling with a cryoprobe [32]. In a retrospective study by Jacob et al., they were able to demonstrate a diagnostic yield of up to 92.6% on a small sample size in the study [33]. The main advantage of TBLC is related to larger biopsy sample in comparison to traditional transbronchial biopsy with associated crushing artifact of the sample. However, TBLC has the limitations related to training and the need for general anesthesia. Moreover, there is a significant risk of complications including pneumothorax and bleeding ranging from 9 to 10% and from 14 to 20%, respectively [34].

Hagmeyer et al. demonstrated that the risk of severe complications can be reduced from 84 to 14% by technical modification of this procedure. Hence, it may prove to be a useful step before consideration of surgical lung biopsy [35].

2.3 Ultrasound (US)-guided biopsy

The diagnostic workup for sarcoidosis should include the least invasive investigation at the outset of evaluation; thus, neck ultrasound may provide an ideal modality for that purpose. We have demonstrated that a core biopsy of cervical lymph nodes (ranging from 7 to 14 mm in diameter), with no sonographic appearance of being marked was adequate to make a histological diagnosis of sarcoidosis [36]. In this retrospective analysis, we showed that if there were no suitable cervical lymph node for biopsy, an US evaluation and biopsy of abnormal parotid glands may help establish the diagnosis of sarcoidosis [36]. In view of the ease of this procedure and its cost-effectiveness (approximately £1000/= saving in comparison to EBUS-TBNA per patient in the UK), this could potentially be considered as a first-line investigation if appropriate expertise is available. Hence, it is proposed that the diagnostic algorithm as shown in **Figure 5** for the investigation of mediastinal lymphadenopathy should include US-guided core biopsy of cervical lymph nodes +/− parotid glands if deemed abnormal. We envisage that a prospective multicenter study of wider application of this technique would be desirable to generalize the use of this minimally invasive diagnostic modality.

2.4 Surgical techniques: cervical mediastinoscopy and video-assisted thoracoscopy (VATS)

2.4.1 Cervical mediastinoscopy

Cervical mediastinoscopy (CM) is one of the preferred surgical techniques to evaluate mediastinal lymphadenopathy. The procedure helps to obtain samples from paratracheal and subcarinal lymph nodes. Since the advent of EBUS-TBNA, EUS-FNA, and PET-CT, the frequency of mediastinoscopy as a diagnostic procedure has declined significantly. Moreover, it is reserved when the above techniques have been inconclusive or not feasible due to the location of the enlarged lymph node.

Onat et al. reported the safety of CM and demonstrated that it is a reliable method, in the evaluation of mediastinal lymphadenopathy [37]. In a meta-analysis by Agarwal et al., they reported a diagnostic yield between 82 and 97% for cervical mediastinoscopy [38].

2.4.2 Video-assisted thoracoscopic surgery (VATS)

VATS biopsy should be considered when there is difficulty in establishing the diagnosis with other less invasive options especially if a specific histological diagnosis would help the prognosis or treatment [39]. The diagnostic yield, sensitivity,

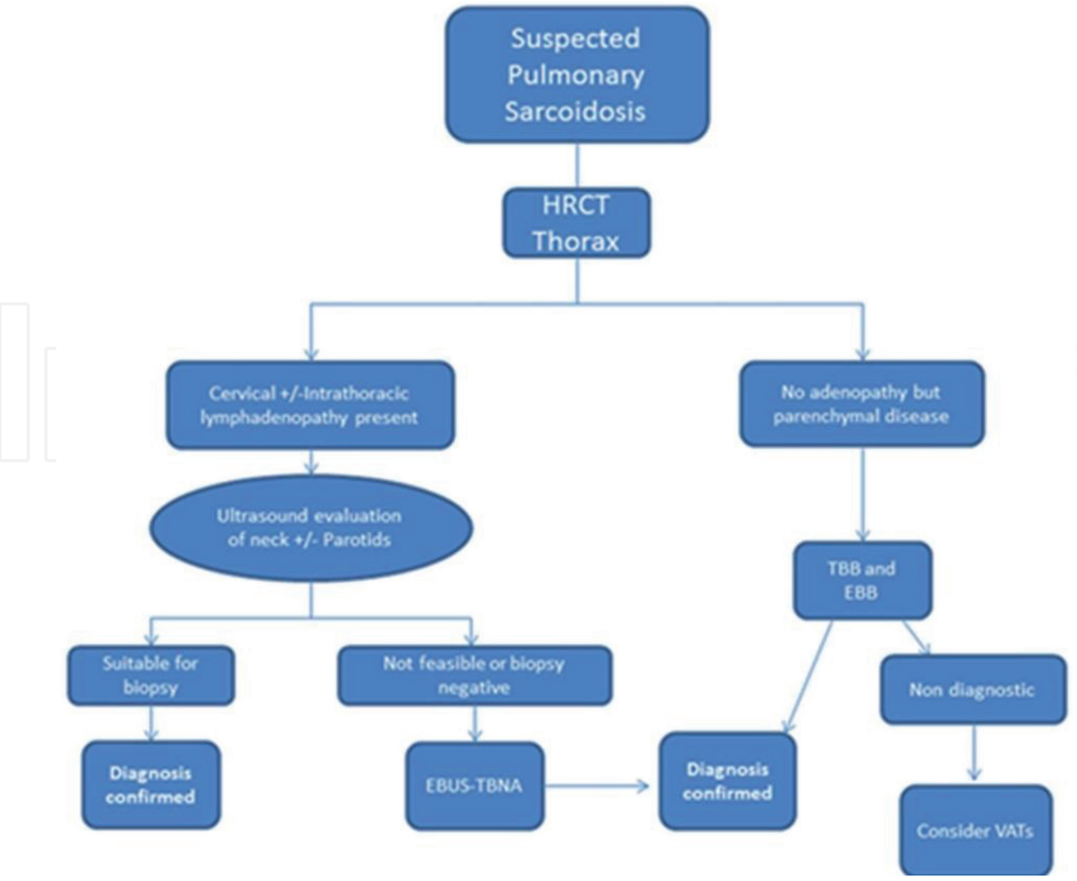

Figure 5.
Diagnostic approach in suspected pulmonary sarcoidosis. EBUS-TBNA, endobronchial ultrasound-guided transbronchial needle aspiration; TBB, transbronchial biopsy; EBB, endobronchial biopsy; VATS, video-assisted thoracoscopic biopsy [36].

and specificity of VATS reported in a meta-analysis were 92.7% (87.6–95.8%), 91% (89–92%), and 58% (31–81%), respectively [40]. VATS procedures have the advantage of technically enabling multilobed biopsies in comparison to open lung biopsy [41]. Currently, mini-VATS is also being increasingly considered given the less postoperative complications and decreased length of hospital stay [42, 43]. VATS procedure should be done by experienced thoracic surgeons, as there is a potential need for mini-thoracotomy in 25% of cases, to obtain adequate tissue for diagnosis [44]. Furthermore, there is a mortality rate of approximately 2% at 30 days associated with this surgical procedure as demonstrated by a meta-analysis conducted by Wallis et al. [45].

3. Conclusion

Sarcoidosis presents as a diagnostic dilemma in a number of medical specialties ranging from pulmonology, general internal medicine, rheumatology, and oncology to name a few. The advent of ultrasound-guided techniques (EBUS-TBNA, EUS-TBNA, and US-guided core biopsy of neck nodes) has significantly reduce the frequency of more invasive diagnostic procedures such as mediastinoscopy and surgical lung biopsies (both open and VATS biopsies). Moreover, TBB is rarely required to sample the lung parenchyma as the diagnostic yield of alternative procedures, with much less associated risk of pneumothorax (EBUS, EUS and US-guided core biopsy of neck nodes), is very high in appropriate clinical context. TBLC may be a newer diagnostic intervention being utilized in selected centers for histological

assessment of ILDs, but its role in the diagnostic pathway is uncertain at present. However, future studies may shed light on its value in the diagnostic pathway of ILD. It is proposed that expertise for US-guided neck node core biopsy would be an important adjunct in the armory of interventional radiologists skill sets and has the potential to be a safe and cost-effective procedure in suspected pulmonary sarcoidosis. Furthermore, learning this technique to sample near normal-sized lymph nodes would be appropriate in minimizing bronchoscopic procedures, preventing significantly higher morbidity.

Acknowledgements

We thank Mr. John Hudson, the librarian at the New Cross Hospital, for his help with the literature review for this chapter.

Conflict of interest

The authors declare no conflict of interest in relation to this manuscript.

Author details

Rajarajan Anandavelu and Ahmed Fahim*
New Cross Hospital, Wolverhampton, UK

*Address all correspondence to: ahmedfahim@doctors.org.uk

References

[1] Calandriello L, Walsh SLF. Imaging for sarcoidosis. Seminars in Respiratory and Critical Care Medicine. 2017;**38**(4):417-436

[2] Drent M, De Vries J, Lenters M, Lamers RJ, Rothkranz-Kos S, Wouters EF, et al. Sarcoidosis: Assessment of disease severity using HRCT. European Radiology. 2003;**13**(11):2462-2471

[3] Ganeshan D, Menias CO, Lubner MG, Pickhardt PJ, Sandrasegaran K, Bhalla S. Sarcoidosis from head to toe: What the radiologist needs to know. Radiographics. 2018;**38**(4):1180-1200

[4] De Boer S, Milne DG, Zeng I, Wilsher ML. Does CT scanning predict the likelihood of a positive transbronchial biopsy in sarcoidosis? Thorax. 2009;**64**(5):436-439

[5] Mostard RL, van Kroonenburgh MJ, Drent M. The role of the PET scan in the management of sarcoidosis. Current Opinion in Pulmonary Medicine. 2013;**19**(5):538-544

[6] Yakar A, Yakar F, Sezer M, Bayram M, Erdogan EB, Ozkan D, et al. Use of PET-CT for the assessment of treatment results in patients with sarcoidosis. Wiener Klinische Wochenschrift. 2015;**127**(7-8):274-282

[7] Keijsers RG, Verzijlbergen EJ, van den Bosch JM, Zanen P, van de Garde EM, Oyen WJ, et al. 18F-FDG PET as a predictor of pulmonary function in sarcoidosis. Sarcoidosis, Vasculitis, and Diffuse Lung Diseases. 2011;**28**(2):123-129

[8] Maturu VN, Rayamajhi SJ, Agarwal R, Aggarwal AN, Gupta D, Mittal BR. Role of serial F-18 FDG PET/CT scans in assessing treatment response and predicting relapses in patients with symptomatic sarcoidosis. Sarcoidosis, Vasculitis, and Diffuse Lung Diseases. 2016;**33**(4):372-380

[9] Teirstein AS, Machac J, Almeida O, Lu P, Padilla ML, Iannuzzi MC. Results of 188 whole-body fluorodeoxyglucose positron emission tomography scans in 137 patients with sarcoidosis. Chest. 2007;**132**(6):1949-1953

[10] Yu C, Xia X, Qin C, Sun X, Zhang Y, Lan X. Is SUVmax helpful in the differential diagnosis of enlarged mediastinal lymph nodes? A Pilot Study. Contrast Media & Molecular Imaging. 2018;**2018**:3417190

[11] Viles-Gonzalez JF, Pastori L, Fischer A, Wisnivesky JP, Goldman MG, Mehta D. Supraventricular arrhythmias in patients with cardiac sarcoidosis prevalence, predictors, and clinical implications. Chest. 2013;**143**(4):1085-1090

[12] Smedema JP, van Geuns RJ, Ector J, Heidbuchel H, Ainslie G, Crijns H. Right ventricular involvement and the extent of left ventricular enhancement with magnetic resonance predict adverse outcome in pulmonary sarcoidosis. ESC Heart Failure. 2018;**5**(1):157-171

[13] Fahim A, Mann JS. Pulmonary sarcoidosis: Diagnostic and treatment update. Expert Review of Respiratory Medicine. 2014;**8**(4):493-501

[14] Hsu RM, Connors AF Jr, Tomashefski JF Jr. Histologic, microbiologic, and clinical correlates of the diagnosis of sarcoidosis by transbronchial biopsy. Archives of Pathology & Laboratory Medicine. 1996;**120**(4):364-368

[15] Shorr AF, Torrington KG, Hnatiuk OW. Endobronchial biopsy for sarcoidosis: A prospective study. Chest. 2001;**120**(1):109-114

[16] Mitchell DM, Mitchell DN, Collins JV, Emerson CJ. Transbronchial lung biopsy through fibreoptic bronchoscope in diagnosis of sarcoidosis. British Medical Journal. 1980;**280**(6215):679-681

[17] Haponik EF, Summer WR, Terry PB, Wang KP. Clinical decision making with transbronchial lung biopsies. The value of nonspecific histologic examination. The American Review of Respiratory Disease. 1982;**125**(5):524-529

[18] Wall CP, Gaensler EA, Carrington CB, Hayes JA. Comparison of transbronchial and open biopsies in chronic infiltrative lung diseases. The American Review of Respiratory Disease. 1981;**123**(3):280-285

[19] Poletti V, Patelli M, Ferracini R, Simonetti M, Spiga L. Transbronchial lung biopsy in infiltrative lung disease. The importance of the pathologic approach. Sarcoidosis. 1988;**5**(1):43-50

[20] Thillai M, Chang W, Chaudhuri N, Forrest I, Ho LP, Lines S, et al. Sarcoidosis in the UK: Insights from British Thoracic Society registry data. BMJ Open Respiratory Research. 2019;**6**(1):e000357

[21] Muller-Quernheim J, Saltini C, Sondermeyer P, Crystal RG. Compartmentalized activation of the interleukin 2 gene by lung T lymphocytes in active pulmonary sarcoidosis. Journal of Immunology. 1986;**137**(11):3475-3483

[22] Prasse A, Georges CG, Biller H, Hamm H, Matthys H, Luttmann W, et al. Th1 cytokine pattern in sarcoidosis is expressed by bronchoalveolar CD4+ and CD8+ T cells. Clinical and Experimental Immunology. 2000;**122**(2):241-248

[23] Tanriverdi H, Erboy F, Altinsoy B, Uygur F, Arasli M, Ozel Tekin I, et al. Bronchoalveolar lavage fluid characteristics of patients with sarcoidosis and nonsarcoidosis interstitial lung diseases: Ten-year experience of a single center in Turkey. Iranian Red Crescent Medical Journal. 2015;**17**(10):e31103

[24] Meyer KC, Raghu G, Baughman RP, Brown KK, Costabel U, du Bois RM, et al. An official American Thoracic Society clinical practice guideline: The clinical utility of bronchoalveolar lavage cellular analysis in interstitial lung disease. American Journal of Respiratory and Critical Care Medicine. 2012;**185**(9):1004-1014

[25] Garwood S, Judson MA, Silvestri G, Hoda R, Fraig M, Doelken P. Endobronchial ultrasound for the diagnosis of pulmonary sarcoidosis. Chest. 2007;**132**(4): 1298-1304

[26] Kitamura A, Takiguchi Y, Kurosu K, Takigawa N, Saegusa F, Hiroshima K, et al. Feasibility of cytological diagnosis of sarcoidosis with endobronchial US-guided transbronchial aspiration. Sarcoidosis, Vasculitis, and Diffuse Lung Diseases. 2012;**29**(2):82-89

[27] Oki M, Saka H, Kitagawa C, Kogure Y, Murata N, Ichihara S, et al. Prospective study of endobronchial ultrasound-guided transbronchial needle aspiration of lymph nodes versus transbronchial lung biopsy of lung tissue for diagnosis of sarcoidosis. The Journal of Thoracic and Cardiovascular Surgery. 2012;**143**(6):1324-1329

[28] Von Bartheld MB, Dekkers OM, Szlubowski A, Eberhardt R, Herth FJ, in 't Veen JC, et al. Endosonography vs conventional bronchoscopy for the diagnosis of sarcoidosis: The GRANULOMA randomized clinical trial. Journal of the American Medical Association. 2013;**309**(23):2457-2464

[29] Gupta D, Dadhwal DS, Agarwal R, Gupta N, Bal A, Aggarwal AN.

Endobronchial ultrasound-guided transbronchial needle aspiration vs conventional transbronchial needle aspiration in the diagnosis of sarcoidosis. Chest. 2014;**146**(3):547-556

[30] Plit ML, Havryk AP, Hodgson A, James D, Field A, Carbone S, et al. Rapid cytological analysis of endobronchial ultrasound-guided aspirates in sarcoidosis. The European Respiratory Journal. 2013;**42**(5):1302-1308

[31] Michael H, Ho S, Pollack B, Gupta M, Gress F. Diagnosis of intra-abdominal and mediastinal sarcoidosis with EUS-guided FNA. Gastrointestinal Endoscopy. 2008;**67**(1):28-34

[32] Pedro C, Melo N, Novais EBH, Magalhaes A, Fernandes G, Martins N, et al. Role of bronchoscopic techniques in the diagnosis of thoracic sarcoidosis. Journal of Clinical Medicine. 2019;**8**(9):1327. DOI: 10.3390/jcm8091327

[33] Jacob M, Bastos HN, Mota PC, Melo N, Cunha R, Pereira JM, et al. Diagnostic yield and safety of transbronchial cryobiopsy in sarcoidosis. ERJ Open Research. 2019;**5**(4):00203-2019. DOI: 10.1183/23120541.00203-2019

[34] Shafiq M, Lee H, Yarmus L, Feller-Kopman D. Recent advances in interventional pulmonology. Annals of the American Thoracic Society. 2019;**16**(7):786-796

[35] Hagmeyer L, Theegarten D, Wohlschlager J, Hager T, Treml M, Herkenrath SD, et al. Transbronchial cryobiopsy in fibrosing interstitial lung disease: Modifications of the procedure lead to risk reduction. Thorax. 2019;**74**(7):711-714

[36] Fahim A, Qasim MM, Rosewarne D. Neck as mediastinal extension: Diagnosis of sarcoidosis by core biopsy of cervical lymph nodes. The Clinical Respiratory Journal. 2020;**14**(1):16-20. DOI: 10.1111/crj.13094

[37] Onat S, Ates G, Avci A, Yildiz T, Birak A, Akgul Ozmen C, et al. The role of mediastinoscopy in the diagnosis of non-lung cancer diseases. Therapeutics and Clinical Risk Management. 2017;**13**:939-943

[38] Agarwal R, Srinivasan A, Aggarwal AN, Gupta D. Efficacy and safety of convex probe EBUS-TBNA in sarcoidosis: A systematic review and meta-analysis. Respiratory Medicine. 2012;**106**(6):883-892

[39] Deconinck B, Verschakelen J, Coolen J, Verbeken E, Verleden G, Wuyts W. Diagnostic workup for diffuse parenchymal lung disease: Schematic flowchart, literature review, and pitfalls. Lung. 2013;**191**(1):19-25

[40] Iftikhar IH, Alghothani L, Sardi A, Berkowitz D, Musani AI. Transbronchial lung cryobiopsy and video-assisted thoracoscopic lung biopsy in the diagnosis of diffuse parenchymal lung disease. A meta-analysis of diagnostic test accuracy. Annals of the American Thoracic Society. 2017;**14**(7):1197-1211

[41] Bradley B, Branley HM, Egan JJ, Greaves MS, Hansell DM, Harrison NK, et al. Interstitial lung disease guideline: The British Thoracic Society in collaboration with the Thoracic Society of Australia and New Zealand and the Irish Thoracic Society. Thorax. 2008;**63**(Suppl 5):v1-v58

[42] Jeon CS, Yoon DW, Moon SM, Shin S, Cho JH, Lee SM, et al. Non-intubated video-assisted thoracoscopic lung biopsy for interstitial lung disease: A single-center experience. Journal of Thoracic Disease. 2018;**10**(6):3262-3268

[43] Massone PP, Lequaglie C, Magnani B, Ferro F, Cataldo I. The real impact and usefulness of video-assisted

thoracoscopic surgery in the diagnosis and therapy of clinical lymphadenopathies of the mediastinum. Annals of Surgical Oncology. 2003;**10**(10):1197-1202

[44] Hsu CP, Hanke I, Douglas JM Jr. Diagnostic video-assisted thoracoscopic procedures. Annals of Surgery. 1995;**222**(5):626-631

[45] Wallis A, Spinks K. The diagnosis and management of interstitial lung diseases. BMJ. 2015;**350**:h2072