The use of auto-antibody testing in the evaluation of interstitial lung disease (ILD) – A practical approach for the pulmonologist

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A B S T R A C T

Interstitial lung diseases (ILD), also defined as diffuse parenchymal lung diseases (DPLD) include a heterogeneous group of pulmonary disorders. They may be caused by an underlying connective tissue disease (CTD), Rheumatoid Arthritis (RA) or ANCA-associated Vasculitis (AAV). Pulmonary manifestations of these conditions may also precede systemic onset and therefore, pulmonologists may be confronted with diagnosing a systemic rheumatic disease. For the discrimination of CTD-related ILD and idiopathic interstitial pneumonia (IIP), serological testing is recommended. After careful reviewing the available literature, we suggest a serologic diagnostic algorithm for pulmonologists dealing with ILD-patients. This algorithm depicts the consensus for antibody testing that was reached amongst authors. Obviously this consensus approach requires further validation in everyday practice and leaves room for local adaption of the diagnostic strategy depending on the availability of diagnostic capacity and cost. It is our hope, however, that the rational and stepwise approach of serological testing for ILD will ultimately save unnecessary expenses associated with general laboratory screening. Finally a broader consensus on the strategy for laboratory testing in ILD in general might also improve the detection level of these relatively rare diseases and this will ultimately improve management and care of patients suffering from these complex disorders.

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1. Introduction

Interstitial lung disease (ILD), also defined as diffuse parenchymal lung disease (DPLD) include a heterogeneous group of pulmonary diseases, sharing several clinical, functional, radiologic and pathologic similarities [1–3] but also presenting with many differences [4], including prognosis and response to treatment. ILDs are usually classified on the basis of aetiology, being defined as “idiopathic” [5], or secondary to known causes, e.g. connective tissue disease, infections, drugs and radiation-induced lung disease or occupational and environmental exposure.

One of the main known causes of ILDs is an underlying connective tissue disease (CTD) [5], or ANCA-associated Vasculitis (AAV) [6]. The pulmonary manifestation of a connective tissue disease or vasculitis can precede the systemic onset [7,8]. Therefore, the physician being primarily involved in diagnosing a systemic rheumatic disease might be a pulmonologist [9]. Recognition of an underlying CTD is particularly challenging when ILD is its first or lone manifestation [10]. The discrimination is important, as the prognosis of patients with connective tissue disease-related ILD (CTD-ILD) tends to be less severe than that of idiopathic ILDs [11–13] - although still controversial [14] - and therapeutic options are different.

In clinical practice, pulmonologists dealing with ILD patients are confronted with patients presenting with subtle clinical features suggesting an underlying autoimmune process, though not meeting established criteria for a distinct connective tissue disease (CTD). Criteria and terms to describe this subset of patients differed between various research groups: “undifferentiated CTD-associated ILD” (UCTD-ILD) [15], "lung-dominant CTD" [16] or "autoimmune-featured ILD" [17]. Recently, an ERS/ATS Task Force introduced the concept and definition of “interstitial pneumonia with autoimmune features” (IPAF) [18], offering harmonized classification criteria based on clinical, serological and morphological features. This consensus approach is important to raise the awareness for this patient subgroup, to uniform clinical approach and to guide future research — recommendations for diagnostic testing, clinical care and management of patients, however, may not yet be derived from this concept and are still left to the individual health care provider [18].

The current international guidelines on the diagnosis of IPF recommend excluding the presence of CTD [19], which inevitably should involve the assessment of extra-thoracic features of CTD (medical history and physical examination by a dedicated physician), testing for circulating auto-antibodies and integrating radiological and histopathological findings [18,19]. However, the diagnostic value of autoantibody testing in excluding connective tissue disease-related ILD, remains sometimes unclear and recommendations regarding the significance of the single auto-antibody tests or the necessity to repeat auto-antibody testing after some time are lacking. Especially the emerging field of antisyntetase autoantibodies is challenging for an evidence-based diagnostic approach.

In this article, we therefore suggest a diagnostic algorithm for autoantibody testing, based on a current literature screen. This algorithm shall give the pulmonologist, being first contacted by a patient with ILD of unknown origin, a pragmatic and clinically orientated code of practice for serological testing. While autoantibodies are fundamental diagnostic tools, it is important to emphasize that auto-antibody results do not definitively prove or disprove a disease. This algorithm is meant to assist current diagnostic guidelines on the detection of ILD and CTD, which may be found elsewhere. We believe that the use of such an algorithm in daily clinical practice can be associated with a reduction in the number of second-level tests and a higher cost-effectiveness in laboratory testing [20,21]. Furthermore, there is a growing appreciation that a multidisciplinary approach to CTD-ILDs can further enhance the diagnostic performance [22].

2. Auto-antibody tests and their clinical significance

2.1. Antinuclear antibodies — ANA

The most commonly used technique for ANA determination is indirect immunofluorescence (IIF). The antigens of Human Epithelial cell line-2 (HEp-2), derived from human larynx carcinoma cells, are used to detect relevant autoantibodies [23]. The ANA-test by immunofluorescence is preferred as screening method because of its high sensitivity [25]. The cut-off for positive test results is suggested to be 1:160 [24], although this cannot be generalized because of the high variability of the test itself and local, technician-dependant differences. High autoantibody titers (>1:160) can be detected in sick subjects, but in only 5% of healthy people [25]. Low antibody titers (1:40) can be detected in about 25–30% of healthy controls [26] and should therefore be considered as negative test result. The frequency of unspecified ANA-positivity in [27] healthy controls seems to increase with increasing age [26]. In a study by Tan et al. (1997), however, the frequency of ANA positivity in healthy subjects did not differ significantly across the 4 age subgroups spanning 20–60 years of age [25].

The autoantibodies detected in the ANA-test are generally referred to as antinuclear antibodies owing to their predominant reactivity with nuclear antigens. However, the ANA-test also detects some antigens that are located in the cytoplasm [28].

The type of distribution pattern observed in IIF on human epithelial cell line-2 (HEp-2) cells in interphase is correlated with antibody specificity, and the signals can originate from nuclear, mitotic or cytoplasmatic domains [28] (see Table 1). Anti-Ro(SSA) and anti- r-T-RNA synthetases, however, are not readily detected owing to the scarcity of antigen in HEp-2 cells and/or leaching and denaturation secondary to fixation procedures [28,31].

The ANA-test is performed in the majority of patients with an ILD of unknown origin in order to exclude an underlying CTD [29]. ANA-positivity at baseline visit may be up to 56% in patients with unclear ILD and performed ANA-test [30]. The probability to
develop an overt CTD is significantly higher in initially ANA-positive patients compared to initially ANA-negative patients, and in younger patients compared to elderly patients [29,30]. A negative ANA-test, however, should be repeated at a future time, as the test might turn positive with change of the clinical course and proceeding time [31]. Furthermore, even a negative ANA-test may justify further rheumatologic evaluation in case of a suspicious clinical picture. The range of newly diagnosed CTDs in patients referred to a specialized clinic for evaluation of unclear ILD varies widely (3.8–19%) in several studies [7,29,30].

The most common types of subsequently diagnosed CTDs are Rheumatoid Arthritis, inflammatory myositis and undifferentiated connective tissue disease (UCTD) [7,30]. As mentioned above, ILDs related to SSA and t-RNA-synthetase antibodies (Sjögren Syndrome, Idiopathic Inflammatory Myositis, Anti-Synthetase Syndrome), can be diagnosed despite ANA-negativity. Therefore, myositis may be underrecognized in the ILD-population, as levels of CK and aldolase are often not markedly elevated, ANA-negativity is possible and amyopathic forms with isolated lung disease do exist [30].

2.1.1. t-RNA synthetase (ARS-) antibodies: inflammatory myopathy and ASS

Cytoplasmatic fluorescence is a typical pattern for anti-t-RNA synthetase antibodies in IIF on HEP2 cells [28]. However, due to the scarcity of antigen in HEP-2 cells and the fixation procedures of these cells, the associated autoantibodies are not always detected [28]. Cytoplasmatic fluorescence in the ANA-test may only be present in less than 50% of patients with positive t-RNA synthetase antibodies [32].

The t-RNA synthetase antibodies belong to a group of antibodies, classified as myositis-specific. Myositis-specific antibodies can be found in approximately 20–40% of patients with idiopathic inflammatory myositis [dermatomyositis (DM), polymyositis (PM), inclusion body myositis] [33–36]. Among the t-RNA synthetase antibodies, anti-Jo-1 antibody is the most common antibody associated with the antisynthetase syndrome, since it is found in approximately 20–30% of patients with PM and 2–10% of those with DM [36–39]. In patients with myositis and ILD, anti-Jo-1 antibodies are detected in about 30–50% [39–41].

Other t-RNA synthetase antibodies, namely the anti-PL-7, anti-PL-12, anti-OJ, anti-EJ and anti-KS might be detected, all together in no more than 10% of patients with myositis [34,42]. The anti EJ antibody seems to be more frequently associated with cutaneous manifestations of DM, while the anti-PL 7 and anti-PL 12 seem to be more frequently associated with the presence of pulmonary fibrosis in the absence of myositis [43]. Regarding the OJ and KS antibodies, clear recommendations concerning the use of these antibodies in patients with ILD are hardly possible because of the low frequency and the low level of evidence.

Interstitial lung disease is commonly found in patients with myositis and especially those with positive anti-synthetase antibodies [44–46]. The prevalence of ILD varies widely (40–80%) among case series of patients with polymyositis (PM) and dermatomyositis (DM) [47–49] and is a significant cause of mortality in PM/DM [50]. Patients with new-onset and unclear ILD may or may not display signs of myositis or skin disease right from the beginning, and the lack of correlation between muscle disease and pulmonary symptoms often leads to delayed diagnosis and a compromised therapeutic response [49,51]. A special entity of myositis, often lacking muscle weakness, while other systemic features, like lung disease predominate, is the anti-synthetase syndrome (ASS) [52]. Anti-t-RNA synthetase autoantibodies are the hallmarks of the ASS, which is characterized by multiple organ involvement, primarily ILD, and is often accompanied by myositis, non-erosive arthritis, Raynaud’s phenomenon, “mechanic’s hands”, skin rashes, sicca syndrome and constitutional symptoms, such as fever [53,54] (see Table 2). Not all patients with anti-synthetase antibodies or even those classified as having the anti-synthetase syndrome have all manifestations of this syndrome. The prevalence of interstitial lung disease in ASS is reported as 65–90% in some studies [55–57]. The existence of interstitial lung disease and absence of myositis is seen more often with some anti-synthetase antibodies than with others [56]. This is why different phenotypes within the anti-synthetase syndrome can be assumed [58].

As mentioned above, anti Jo-1 is the most common autoantibody whereas autoantibodies other than Jo-1 are found altogether in 1–7% of these patients. Among non-Jo-1 ASS, anti PL-7 and anti PL-12 autoantibodies appear to be most common [59–61], although reliable figures are missing. The prevalence of ILD in anti-

Table 2
Anti-synthetase syndrome.

<table>
<thead>
<tr>
<th>Antibody</th>
<th>CTD</th>
</tr>
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<tbody>
<tr>
<td>dsDNA</td>
<td>SLE</td>
</tr>
<tr>
<td>U1-RNP</td>
<td>MCTD</td>
</tr>
<tr>
<td>Ro/SSA, La/SSB</td>
<td>Sjögren Syndrome</td>
</tr>
<tr>
<td>SmD</td>
<td>SLE</td>
</tr>
<tr>
<td>Mi-2</td>
<td>Myositis (DM)</td>
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<tr>
<td>CENP-B</td>
<td>SSC</td>
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<tr>
<td>RNA-Polymerase III</td>
<td>SSC</td>
</tr>
<tr>
<td>Topoisomerase 1 (SCL70)</td>
<td>SSC</td>
</tr>
<tr>
<td>Th/To</td>
<td>Overlap Syndrome</td>
</tr>
<tr>
<td>PM-SCL 75/100</td>
<td>ASS/Myositis</td>
</tr>
<tr>
<td>t-RNA synthetase (Jo-1, PL-7, PL-12)</td>
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</tr>
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</table>

Table 1
ANA fluorescence pattern and related auto-antibodies.

<table>
<thead>
<tr>
<th>ANA Pattern</th>
<th>Antibody</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nuclear (most frequent)</td>
<td>- homogeneous</td>
</tr>
<tr>
<td></td>
<td>- speckled</td>
</tr>
<tr>
<td></td>
<td>- centromeric</td>
</tr>
<tr>
<td></td>
<td>- nucleolar</td>
</tr>
<tr>
<td>Cytoplasmatic (less frequent)</td>
<td>- speckled</td>
</tr>
<tr>
<td>Mitotic (rare)</td>
<td>- mitochondrial, ribo-somal, cytoskelatal, Golgi apparatus, lysosomes</td>
</tr>
</tbody>
</table>

ANA fluorescence pattern and related specific auto-antibodies, such as underlying CTD [1,28,54], ASS: Anti-synthetase syndrome. MCTD: Mixed Connective Tissue Disease. SLE: Systemic Lupus Erythematodes. SSC: Systemic Sclerosis.
PL-7 positive patients varies from 77 to 100% [60,62]. Other symptoms of ASS seem less common in patients with anti-PL-7 antisynthetase syndrome [62]. In a study with patients having PL-12 antibodies, 90% had ILD, 90% were diagnosed for an underlying CTD and 65% were primarily referred to a pneumologist [9].

Non-Jo-1 synthetase antibodies like anti PL-7 and anti PL-12 might define a phenotype that is distinct from that of anti-Jo-1-positive patients and is characterized by a lower incidence of myositis and a higher incidence of ILD [63]. These antibodies may be more common among patients presenting with “idiopathic” interstitial pneumonia than formerly considered and should be checked in patients with features of antisynthetase syndrome despite a negative screen for anti-nuclear or anti-Jo-1 antibodies [60]. The response to immunosuppressive medications is generally, but not universally favorable [63], underlining the therapeutic relevance.

2.1.2. Mi-2 and CADM-140 antibodies: inflammatory myopathy

Besides the t-RNA-synthetase antibodies, there are further antibodies described as myositis-specific, namely antibodies to signal recognition particle (SRP), and to the nuclear helicase Mi-2 [64]. The Mi-2 antibody, was first linked to dermatomyositis in 1985 [65] and has a diagnostic sensitivity and specificity of approximately 4–18% and 98–100%, respectively. In a recent study by Cruellas and colleagues, enrolling 127 patients with dermatomyositis, Mi-2 positive patients were frequently affected by pulmonary involvement [66]. Whether ILD may be the first and single sign of Mi-2 related dermatomyositis is not described in the currently published literature. Screening for Mi-2 may be useful in suspicious patients affected by unclear ILD.

There is, however, a subset of patients with idiopathic inflammatory myopathy that may have dissociated skin and skeletal muscle involvement, and patients may have florid cutaneous manifestations of dermatomyositis for a prolonged period of time - at least 2 years according to the definition [67,68] - without any evidence of underlying myositis. This condition has been termed amyopathic dermatomyositis (ADM or dermatomyositis sine myositis), and has been included in the clinical spectrum of idiopathic inflammatory myopathy [69,70]. It has recently been shown that ILD may be associated with and may antedate amyopathic dermatomyositis [71]. Thus, particular attention should be paid to cutaneous changes in patients with ILD, such as heliotrope rash, facial erythema and oedema, Gottron’s papules, and perungueal telangiectasia, especially if the histopathological pattern is NSIP [72].

The recent identification of antibodies that may be specifically associated with amyopathic dermatomyositis (CADM-140 antibodies, representing clinically amyopathic dermatomyositis, autoantigen of 140 kDa) might aid an earlier diagnosis of this condition [73]. Patients with positive CADM-140 antibodies (also known as MDA-5 antibodies: melanoma differentiation-associated gene 5) have been shown to display a rapidly progressive ILD, compared to anti-t-RNA synthetase positive myositis patients [74]. Furthermore, CADM-140/MDA-5 levels have been shown to indicate response to treatment and disease activity in patients with DM and rapidly progressive ILD [75,76].

2.1.3. PM/Scl-75/100 antibodies: overlap syndrome

Antibodies directed against PM/Scl are a heterogeneous family of molecules, the most clinically significant of which are PM/Scl-75 and PM/Scl-100. These antibodies give a nucleolar pattern of immunofluorescence at IIF. Both PM/Scl-75 and PM/Scl-100 identify overlap syndromes of Systemic sclerosis and Polymyositis, and give some information about clinical subsets and prognostic details. Their presence has been described as a risk factor for ILD with a high level of specificity for ILD (up to 94.8% for PM/Scl-75 and up to 95.2% for PM/Scl-100.) In a patient with ILD of unknown origin, especially in the presence of a ANA test positivity with nucleolar pattern and a suspicious clinical picture, the detection of both PM/Scl-75 and PM/Scl-100 suggests the diagnosis of overlap syndrome vs. another defined major connective tissue disease [77-79]. A positive ANA-titre and distinct fluorescence pattern might give a first clue into this direction. However, as the clinical appearance may be quite similar to t-RNA-positive ILD patients, the detection of PM/Scl also seems reasonable in case of ANA negativity.

2.1.4. SSA/SSB – antibodies: Sjogren Syndrome

Antibodies against Ro/SSA and La/SSB autoantigens are generally characterized by a nuclear staining pattern with speckled fluorescence in the ANA test [28].

Serum containing autoantibodies directed against the Ro/SSA antigens may recognize one or both of two cellular proteins with molecular weights of approximately 52 and 60 kD (referred to as Ro52 and Ro60) [80,81]. Patients in whom the only autoantibodies present are anti-Ro antibodies may sometimes have a falsely negative ANA test using the traditional human epithelial cell line-2 (HEp-2) cell substrate, because Ro60 immunoreactivity may be lost during denaturation [82]. The detection of Ro/SS-A by immuno-fluorescence, on the other hand, may be missed in the presence of high titre ANAs. Furthermore, considering a detection sensitivity of 91%, a negative immunofluorescence result for Ro/SS-A does not exclude the presence of this autoantibody [83]. Although La/SSB has been shown to shuttle between the nucleus and cytoplasm, the protein is predominantly found in the nucleus of HEp-2 cells. Autoantibodies directed against La produce a speckled nuclear staining pattern and are correlated to internal organ dysfunction (e.g. lung) [84].

Both SSA and SSB antibodies primarily identify patients with Sjögren Syndrome and Systemic Lupus Erythematosus (SLE). SSA antibodies may further be present in other autoimmune diseases, including systemic sclerosis, idiopathic inflammatory myopathies and rheumatoid arthritis. In dermatomyositis, the presence of anti-Ro-52 antibody is correlated with pulmonary disorders [66]. In contrast to anti-Ro/SSA antibodies, anti-La/SSB antibodies are relatively specific for the diagnosis of Sjögren Syndrome [85].

Having a close look at patients with primary Sjögren Syndrome and interstitial lung disease, the underlying CT-pattern (UIP vs. NSIP) seems to have no influence on disease-progression or prognosis [86]. In general, pulmonary manifestations in Sjögren Syndrome have a slow progression and favorable prognosis, with the exception of primary pulmonary lymphoma and pulmonary hypertension [87]. Furthermore, anti-Ro52/SSA antibodies may occur together with antisynthetase antibodies (Jo-1, PL-7, PL-12) and this combination has been associated with particularly severe ILD [88,89]. Concerning disease-onset, ILD has been shown to be the first and single presentation of patients finally diagnosed with primary Sjögren Syndrome. In this case minor salivary gland biopsy may allow for a more precise diagnosis, besides serological testing, as it is possible that not only ANA and RF are negative but also SS-A and SS-B [90]. For these reasons, excluding Sjögren Syndrome in patients with unclear ILD is not trivial and relies on a thorough medical history, proper physical examination, and comprehensive serological testing, including both ANA and SS-A/SS-B.

2.1.5. Topoisomerase I (topo I/SCL-70), RNA polymerase III, Th/To and CENP-B – antibodies: systemic sclerosis

Systemic sclerosis (SSc) is a systemic autoimmune disease of unknown cause characterized by cutaneous and visceral fibrosis, microvascular obliteration and highly specific serum autoantibodies to nuclear autoantigens [91]. These auto-antibodies may be directed against the centromere Protein B (ACA/anti-CENP-B),...
against topoisomerase I (anti-topo I/SCL-70), against RNA polymerase III or against proteins of the RNase MRP and RNase P ribonucleoprotein complex (Th/To) and are important diagnostic features of systemic sclerosis [91,92]. Altogether, 90% of SSC patients have a positive ANA-test [93] and the four just mentioned specific antibody-tests account for 75–80% of ANA positivity in Systemic Sclerosis [94]. The mere pattern of ANA fluorescence may even distinguish certain specific antibodies: CENP-B antibodies display a distinct centromere pattern [94], whereas anti topo-I, anti RNA-polymerase III and anti Th/To are characterized by a nucleolar fluorescent pattern in the ANA test [94]. The frequency of these antibodies is variable (ACA: 26%, anti topo-I: 28%, anti RNA-polymerase III: 10%, and anti Th/To: 3%) and they are associated with distinct clinical pictures [94].

In clinical terms, a limited cutaneous subset (lcSSc) can be distinguished from a diffuse cutaneous disease (dcSSc). In patients with limited cutaneous SSC, skin sclerosis is restricted to the hands, the distal forearm, such as the face and neck. These patients generally have prominent vascular manifestations, including severe Raynaud phenomenon and cutaneous telangiectasia. Many patients with lcSSc have manifestations of the CREST syndrome (Calcinosis cutis, Raynaud phenomenon, Esophageal dysmotility, Sclerodactyly, and Telangiectasia). Sclerotic skin on the chest, abdomen, or upper arms and shoulders is indicative of diffuse cutaneous SSC and patients are more likely to have significant internal organ damage due to ischemic injury, compared to limited cutaneous SSC. The prevalence of systemic sclerosis related ILD appears to be higher in dcSSC than lcSSC [95]. However, autoantibody status seems to be a more useful predictor of organ involvement, including the presence of lung disease, than the pattern of skin involvement [95].

Anti-centromere antibodies (ACA) are found in 20–30% of SSC patients and CENP-B is the major autoantigen recognized by >95% of ACA-positive sera [94]. In clinical terms, CENP-B antibodies are strongly associated with the CREST syndrome [96]. This CENP-B positive phenotype is frequently and predominantly associated with pulmonary hypertension [97] and a low prevalence or even absence of SSc-ILD [98].

Patients with positive anti-topoisomerase-I antibodies on the other hand are frequently affected by lung fibrosis [99] and appear to be protected against isolated pulmonary arterial hypertension [100]. Topoisomerase I antibodies (also known as SCL-70/anti-scleroderma-70 antibodies) are present in 20–30% of SSC patients, most often with diffuse cutaneous SSC. SSc-ILD is most prevalent in these topoisomerase-positive patients with over 85% developing pulmonary fibrosis [101]. Interestingly, topoisomerase titers seem to correlate with disease severity and activity, both in SSC and SSc-ILD [102,103].

Like anti-topoisomerase-I-positive patients, those with positive RNA-polymerase-III antibodies also cluster with a diffuse form of SSC and restrictive pulmonary involvement [104,92]. However, the clinical condition more frequently influencing the course of these patients is renal crisis [105].

Th/To antibodies only account for 3% of SSc-specific autoantibodies. In one small study, patients, primarily diagnosed as IPF but with positive and nuclear-staining ANA, were subsequently tested for Th/To-antibodies and showed positive results 50% [106]. The role of this rare antibody, however, remains unclear.

The general prevalence of Systemic Sclerosis ranges from 50 to 300/Million [107] and the spectrum of SSc-related ILD ranges from limited and often non-progressive lung involvement, to severe disease which may progress to respiratory failure and death [95]. Raynaud’s phenomenon might be an important first sign of the disease, as it is present in over 90% of all SSc-patients. Although reliable figures are missing, ILD appears to be evident in 55–60% of SSc patients [108] and most of these patients have NSIP as underlying pattern [109]. Detecting SSc-ILD is important in any case, as interstitial lung disease is now the most frequent cause of death in systemic sclerosis, having supplanted renal crisis in that regard [93].

In contrast to the formerly mentioned t-RNA-synthetase antibodies and SSA/SSB antibodies it is unlikely to find SSc-specific auto-antibodies in the presence of a negative ANA-test. Interstitial lung disease is a complication observed relatively late in the course of the disease and hardly as first or lone manifestation [110,111]. Screening for anti-topo-I, Th/To, RNA-polymerase III and CENP-B therefore may be performed just in case of a positive ANA-test and typical clinical signs and symptoms of systemic sclerosis. Furthermore, as SSc-specific auto-antibodies are almost invariably present in high titers extremely early in the disease process [112], they may not necessarily be followed-up, once they have been screened.

2.1.6. Sm and ds-DNA antibodies: systemic lupus erythematoses

In patients with Systemic Lupus Erythematodes, the ANA test is virtually always positive [113]. With respect to the various clinical presentations, there are two autoantibodies that are disease-specific and may therefore ease the diagnostic procedure: ds-DNA and Sm-antibodies. Anti-ds DNA antibodies are specific for SLE, making the presence of these antibodies very useful for distinguishing patients with SLE from patients with other systemic autoimmune diseases and are rarely found in patients with other connective tissue disorders [114]. Antibodies directed against Small Nuclear Ribonucleoprotein Core Proteins (anti-Sm antibodies) are insensitive but also highly specific markers for SLE [115].

Sm-Antibodies are commonly characterized by a speckled fluorescence in the ANA-test. There is some evidence in the literature that ds-DNA antibodies are solely positive with high titres in the ANA-test (>1:6400) [116–118]. Antibodies against dsDNA usually display a homogeneous ANA-pattern [116,119].

Pulmonary problems are common in systemic lupus erythematoses, and may be the presenting feature of this multi-system disease. The clinical spectrum ranges from mild, self-limited, pleuritic chest pain to fulminant and rapidly fatal diffuse pulmonary hemorrhage [120]. The most prevalent respiratory manifestations are pleuritis and pulmonary infections [121]. Interstitial lung disease (ILD) is a minor problem and the clinical progression appears to be slow or may even stabilize with time [122].

Given the above mentioned facts, a patient presenting with unclear ILD as first and lone manifestation of SLE and additionally a negative ANA test is hard to imagine. In order to avoid unnecessary serological testing, it might be reasonable to explicitly search for SLE-specific antibodies in case of (1) a (high) positive ANA-test and (2) suspicious clinical signs and symptoms and a suitable medical history.

2.1.7. U1-RNP antibodies: mixed connective tissue disease (MCTD)

Anti-U1–RNP antibodies are characterized by a speckled nuclear pattern in the ANA-test [28]. Patients with positive U1–RNP and features of more than one specific CTD are described as having mixed connective tissue disease (MCTD), which is a specific CTD itself [123]. MCTD was first described in 1972 as a distinct overlap characterized by features of SSc, SLE and PM/DM, with respiratory involvement occurring in about 80% of patients [123]. Such patients are different from those, who do not fulfill the diagnostic criteria for any specific CTD but display features, including serologic abnormalities and systemic extrapulmonary manifestations, that are merely reminiscent of an underlying autoimmune disease [124]. The latter group of patients can be described as having undifferentiated CTD (UCTD), or in case of predominant ILD: IPAF [18].
While a large number of patients with MCTD have pulmonary involvement, most have relatively mild disease, and many are asymptomatic [125]. Severe lung fibrosis is associated with increased mortality [126]. The most severe clinical manifestation, however, is pulmonary hypertension which contributes to premature death in patients who have MCTD [127].

ANA-testing appears to be positive in virtually all patients with positive U1-RNP antibodies [128]. Therefore it might be eligible to assess U1–RNP antibodies in a patient, presenting with ILD of unclear origin and no signs or symptoms of an underlying CTD just in case of a positive ANA-test.

2.2. ANCA: ANCA-associated vasculitis

Antibodies directed against neutrophil cytoplasmic antigens (ANCA) are associated with different forms of vasculitis. Two types of ANCA assays are currently in wide use: Indirect immunofluorescence assay, using alcohol fixed buffy coat leukocytes and Enzyme-linked immunosorbent assay (ELISA), using purified specific antigens. Of these two techniques, the immunofluorescence assay is more sensitive and the ELISA more specific. The optimal approach to clinical testing for ANCA is therefore to screen with immunofluorescence assays, if available, and to confirm all positive results with ELISA, directed against the vasculitis-specific target antigens [129,130].

The two main patterns of ANCA staining, recognised by indirect immunofluorescence microscopy, are cytoplasmatic (c-ANCA) and perinuclear (p-ANCA). Both target antigens are associated with different clinical syndromes [131]. Perinuclear p-ANCA is most often and almost uniquely associated with syndromes characterized by small vessel vasculitis, such as microscopic polyangiitis (MPA) [132].

ILD is known to be an early lesion of MPA in some cases [133] and positive ANCA-titres in patients presenting with an ILD of unknown origin might be the first sign of ANCA-associated vasculitis [134]. In a number of case reports, retrospective and case-control studies it is reported, that the initial diagnosis of an idiopathic interstitial pneumonia had to be revised during the clinical course on behalf of a newly diagnosed MPA [133–135]. The studies by Nishkantha (2011) and Foulon (2008) contain two important messages. First, virtually all patients displaying a positive ANCA test in the clinical course show a perinuclear pattern (p-ANCA) in IIF, with specificity to MPO in EIA, finally leading to a diagnosis of microscopic polyangiitis. Second, ANCA assessment in many cases was not available at the first manifestation of ILD, which led to a delayed diagnosis.

As clinical, radiologic and bronchoalveolar lavage data are similar between ANCA-positive and ANCA-negative patients at time of diagnosis [135], a systematic screening for ANCA in patients with unclear interstitial lung disease appears reasonable. Screening for p-ANCA may be the first step because of the high sensitivity of IIF. However, p-ANCA can also be positive in other medical conditions and a positive result might be confirmed by screening for MPO-specificity, indicating an underlying microscopic polyangiitis. Nonetheless, about 25% of patients diagnosed as having MPA are ANCA-negative and ANCA status may change over time [136]. A negative ANCA assay at initial screening therefore might be rechecked in the clinical course.

2.3. RF and CCP-antibodies: rheumatoid arthritis

CCP-antibodies have a sensitivity of 57% and a specificity of 96% in detecting patients with rheumatoid arthritis (RA). Compared to rheumatoid factor, CCP-antibodies have greater specificity (96% vs. 86%), with similar sensitivity. There is only insufficient evidence to ascertain whether the combination of anti-CCP and rheumatoid factor provides additional benefit over anti-CCP alone in diagnosing rheumatoid arthritis [137].

The prevalence of rheumatoid arthritis associated ILD (RA-ILD) varies from 1 to 40%, depending on the definition for ILD [138]. A population-based study estimated a prevalence of about 1 in 10 patients with RA [139]. While RA occurs more commonly in females (female to male ratio 3:1), RA-ILD is more frequent in males. The presence of ILD is associated with a significantly shortened survival in patients with rheumatoid arthritis, compared to those patients without ILD, as lung disease is the second most common cause of death for RA patients [138,140]. The manifestation of an ILD can precede the RA diagnosis in some cases [140]. Taking into account that rheumatoid arthritis is the most common of all CTDs, it is reasonable to search for CCP antibodies and RF in patients presenting with an ILD of unknown origin, mainly if the patients present articular symptoms. However, seronegative rheumatoid arthritis is also possible and a negative result of RF and CCP does not exclude an underlying RA. Rechecking RF and CCP in the clinical course of an unclear ILD might be reasonable as the serotype might change over time.

3. Connective tissue disease-related ILD: the pulmonologists point of view

3.1. Epidemiology

To precisely estimate the incidence and prevalence rates of ILD is for several reasons quite difficult. On one hand, before the Consensus Guidelines were enacted in 2002, there was a lack of international standard that has resulted in variable and confusing criteria and nomenclature. On the other hand, despite the guidelines, IIP remains a diagnosis of exclusion requiring extensive investigation, which is seldom possible [141]. Furthermore, many of the available data are taken from registries or hospital clinics and therefore suffer from selection bias [142]. On a population-based approach, there are two important studies.

The most important registry study was undertaken in Bernalillo County, New Mexico, USA. All diffuse interstitial lung diseases together were found to have a prevalence of 60–80/100,000 and an incidence rate around 30/100,000. Sarcoidosis and idiopathic pulmonary fibrosis were the two most frequent diseases, accounting together for more than 50% of all cases, followed by ILD related to connective tissue disease and to immunologic lung disease [143]. The prevalence of ILD in males (80.9/100,000) was in general 20% higher, than in females (67.2/100,000), and the prevalence of preclinical or undiagnosed ILD among all deaths in the New Mexico cohort was found to be 1.8%.

In a Danish nationwide population-based study the overall incidence rate of ILD was around 31 per 100,000 person-years in the time period of 2001–2005 [144]. In general, these results from Kormun et al. in Denmark accord with those from Coulats et al. in New Mexico, despite the use of different case ascertainment methods and patients’ exposure to different environmental factors [144].

Very little data from population-based registries are available from other countries. The few published reports suggest international differences, concerning the prevalence of ILD [141]. The inconsistency of estimated incidence rates may stem from differences in sampling procedures and diagnostic criteria and measurement bias arising from variation in coding practices and case ascertainment [144]. The overall prevalence of connective tissue disease-related ILD among all patients with ILD is estimated to be 7.1%/11.6% (male/female) [145].
3.2. Histopathology

Connective tissue diseases are systemic disorders characterized by autoimmunity and autoimmune-mediated organ damage. The lungs are frequently involved and diffuse parenchymal lung injury patterns, characterized by varying degrees of inflammation and fibrosis, are common manifestations of CTD [8].

In patients with connective tissue disease, NSIP is the most common histological pattern of CTD-ILD [147]. Especially in patients diagnosed as idiopathic NSIP (iNSIP), the development of connective tissue disease, as well as the development of autoimmune thyroiditis has to be considered within the clinical course and might occur in almost 50% of the patients primarily diagnosed iNSIP (11% connective tissue disease, 22% undifferentiated connective tissue disease, 26% autoimmune thyroiditis) [148]. Lung injury often arises within the first two years after disease onset [148] or may be the first manifestation [8], even several months before development of a typical CTD [149]. Therefore, a link between the clinical entity of NSIP and autoimmune disorders is suspected and idiopathic NSIP might represent an early lung manifestation of an autoimmune disease [15,148].

3.3. Prognosis

As mentioned above, NSIP is the histological pattern, seen most often in CTDs - except for RA, characterized by a higher frequency of UIP. Compared to the idiopathic variety of UIP in IPF, UIP pattern in CTD-related ILD is associated with a significantly better survival [150]. Patients not meeting the criteria for a connective tissue disease but having clinical signs and symptoms suggestive of a CTD, suitable serologic tests and/or a suitable morphological pattern (HRCT, Histopathology, multicompartment involvement) may be supposed to have IPAF [18]. The recent consensus definition of this entity surely will raise the awareness within the medical community, but conclusions with respect to prognosis cannot be drawn yet [17].

Rheumatoid Arthritis is by far the most common CTD and clinically significant RA-ILD occurs in about 10% of the Rheumatoid Arthritis population. It has been clearly shown that the presence of RA-ILD is associated with shortened survival and more severe underlying disease [139,151]. Whereas overall mortality rates for RA have fallen, those associated with RA-ILD have increased significantly in older age groups [139]. Better strategies for the treatment of ILD are supposed to lower mortality among individuals with Rheumatoid Arthritis [140].

Concerning the field of t-RNA synthetase antibodies and myositis, variable clinical courses can be observed. Resolution of pulmonary disorders can be detected, as well as deterioration. Mortality rates differ considerably among patients with ILD deterioration and those without [47.1% vs. 3.3%] [152]. Whether more aggressive therapy in PM/DM patients presenting with ILD really leads to a better outcome is still an open question.

3.4. Diagnostic algorithm for serologic autoimmune testing – why?

The main challenge for pulmonologists approaching a patient with HRCT scan evidence of an ILD is to discriminate whether the ILD is idiopathic or secondary to a known cause. As mentioned above, since CTD represents a possible cause of ILD, and ILD may be the first manifestation of some systemic autoimmune disease which can manifest later, an accurate clinical history together with serology must be evaluated.

The main clinical systemic signs and symptoms which should be asked and detected in every patient presenting with an ILD are listed in Table 3.

3.4.1. Prognosis

Furthermore, basic laboratory data should be obtained, including at least complete blood count, indices of inflammation, liver and kidney function, creatine kinase (CK), lactate dehydrogenase (LDH), myoglobin, aldolase, C3 and C4 complement factors, such as serum protein electrophoresis and urine analysis. As listed in the recent ERS/ATS statement [18], a huge number of serologic auto-antibodies represents the serologic domain of classification criteria for IPAF, namely ANA, RF, anti-CCP, anti-ds DNA, anti-Ro (SSA), anti-La (SSB), anti-ribonucleoprotein, anti-Smith, anti-topoisomerase (SCL-70), anti-tRNA synthetase, anti PM-ScI and anti-MDA. Some of these serologic tests are also listed in the ERS/ATS/JRS/ALAT statement on the diagnosis of IPF [19], although recommendations beyond ANA, RF and anti-CCP are vague and irrespective of cost. The predictive value of positive auto-antibody results in the diagnostic approach of unclear ILD remains a matter of scientific debate – and somehow elusive. Positive serologic results may be unspecific laboratory abnormalities, markers of a distinct underlying auto-immune disease, or reflect an ongoing immunologic process that can be described phenomenologically - without the chance of proper etiologic classification, according to current knowledge. Given these considerations and bearing in mind that auto-antibody testing is rather expensive and not easily available all around the globe, this review focuses on two major goals:

1. Evaluating the predictive value of single auto-antibody tests in the context of ILD.
2. Reaching interdisciplinary consensus on a stepwise approach for autoantibody testing in ILD, based on the currently available scientific knowledge, the clinical expertise of dedicated pulmonologists and rheumatologists and also on economic considerations.

So - when to perform serology for autoimmunity? And which serology should be tested in which order? The answer to these questions depends on the clinical presentation, such as the HRCT-pattern and histopathology (if available). According to our opinion, the basic set of laboratory tests for excluding an underlying CTD in the patient presenting without any signs or symptoms reminiscent of an underlying CTD but with a highly probable diagnosis of an idiopathic interstitial pneumonia should meet
the following criteria:

Given the ANA-test as generally accepted screening test, a more comprehensive basic laboratory testing set should include those antibody tests that might produce positive results even in case of ANA-negativity. Furthermore, basic laboratory testing in these patients should also include those antibody tests that identify CTDs, frequently accompanied and preceded by ILD as their first and lone manifestation. The chosen set of laboratory tests should be able to detect frequent errors in the diagnostic work-up. The exclusion of underlying CTD in order to diagnose an idiopathic interstitial pneumonia firstly is the job of a pulmonologist – any positive results in serological evaluation, such as any clinical signs and symptoms indicative of an underlying CTD should, however, lead to a multidisciplinary approach, together with an experienced rheumatologist.

Given the case that medical history, clinical symptoms, HRCT pattern, such as histopathology and the overall impression don’t connect to a conclusive picture – and might now be called IPAF according to 2015 ATS/ERS research statement [18] – a more sophisticated serological testing might be necessary. Especially in middle-aged women with a new diagnosis of ILD and NSIP pattern, a more comprehensive serological testing might be reasonable, since symptoms might be sometimes underestimated, or might be not clear enough for a specific diagnosis. The aim of this advanced laboratory testing is to search for rare cases, rather than excluding the most probable options.

Another important question is, whether an autoimmune assessment should be performed during the follow-up of patients with an existing diagnosis of an idiopathic ILD or IPAF. In our opinion, the answer is positive, especially in patients with NSIP, since evidence demonstrates a later manifestation of a CTD/UCTD in a relevant percentage of cases [148,153]. We suggest the same serologic testing both at baseline and at follow up (e.g. once a year) or whenever the clinical status seems to change.

Although autoimmune thyroiditis does not represent a systemic autoimmune disease, but an organ-specific autoimmune disease, it represents a late autoimmune manifestation in patients with idiopathic NSIP [148], and specific serology might be reasonable as well [TSH, anti-thyroglobulin (anti-Tg), and anti-thyroid peroxidase (anti-TPO) antibodies].

4. Suggestion for a diagnostic algorithm: serological evaluation in patients with ILD

A pragmatic and clinically useful diagnostic algorithm must be able to take several requirements into account: it should be evidence-based, it should be doable in every-day clinical routine and it should be economical. By focusing on a rational set of tests, unnecessary laboratory testing can be avoided and health care costs can be reduced. According to the presented data in the text and after careful reviewing the available literature, we suggest a diagnostic algorithm for pulmonologists dealing with ILD-patients. Fig. 1 depicts the consensus for antibody testing that was reached amongst authors. We recommend that the basic laboratory screening should include tests for autoantibodies against Ro/SSA, La/SSB, Jo-1 and CCP, such as an ANA, ANCA and RF test.

Screening for antinuclear antibodies is generally accepted and a basic diagnostic tool for screening connective tissue diseases with high sensitivity and poor specificity. The pattern of immunofluorescence can provide a first clue which specific antibody-test might give further insight into the exact diagnosis. The probability to detect an underlying connective tissue disease is higher in patients with high ANA titres. Nonetheless, the ANA-test has its limitations, as some specific autoantibodies (tRNA-synthetase, Ro/SSA) are not readily detected. Concerning age-related differences in the ANA-titres, data of elderly control subjects are sparse. Different cutoffs for ANA-positivity are not recommended, but results should be interpreted with caution.

The emerging field of anti-synthetase antibodies and anti-synthetase syndrome is challenging. As there are frequent screening failures in the ANA-test, we recommend including the most common one for myositis – anti Jo-1 – in routine screening. However, if there is reasonable suspicion for myositis, anti-synthetase syndrome or overlap syndrome, further serological testing can be useful, even in case of a negative ANA-test. By current knowledge PL-7 and PL-12 appear to be those antigens, most frequently associated with ILD, and should be part of an advanced laboratory work-up. The assessment of other antibodies like OJ, Ej, KS, Mi-2 and PM/SCL-75/100 depend on the individual circumstances. Idiopathic inflammatory myopathy with ILD as organ complication might be an underrecognised condition and checking especially for the most frequent t-RNA synthetase antibodies might be rewarding.

Ro/SSA and La/SSB antibodies should be included in the routine testing in our opinion, because of relatively frequent screening failures in the ANA test, especially concerning SSA antibodies. Positive results might point to an underlying Sjögren Syndrome, which can be an attendant phenomenon of other CTDs, especially SLE, or can exist as a single specific entity of a connective tissue disease.

Rheumatoid Arthritis is the most common CTD, often associated with ILD. The occurrence of an ILD can precede definite diagnosis of rheumatoid arthritis. Therefore RF and anti-CCP antibody screening should be included in routine screening laboratory testing. In microscopic polyangiitis (MPA), ILD also can be the first apparent symptom, which makes a routine screening for ANCA, especially p-ANCA reasonable and recommendable.

In case of a positive ANA-test and in accordance with the clinical picture, medical history and other diagnostic results, advanced laboratory testing for specific autoantibodies should be considered. Systemic Sclerosis and CREST-Syndrome, Systemic Lupus Erythematoses and Mixed connective tissue disease are often accompanied by clinical, radiologic or other features that might call the physicians attention into a certain direction. For this reason, screening for Scl-70, Th/To, RNA-Polymerase III and CENP-B (SSc/CREST), as well as Sm and dsDNA (SLE) and for U1-RNP (MCTD) might be part of a more sophisticated serological approach in order to search for rare and neglected cases.

Apart from the above mentioned recommendations for serological testing, the importance of medical history and physical examination, such as careful performance and interpretation of radiological images cannot be stressed enough. Even a comprehensive routine laboratory data set might provide first important insights. Screening for viral hepatitis and HIV might also be useful in some cases and should be considered.

The current algorithm aims to combine simplicity and practicality for the work up of pulmonary involvement in ILD. Obviously this consensus approach leaves room for local adaption of the diagnostic strategy depending on the availability of diagnostic capacity and cost. It is our hope, however, that the rational and stepwise approach of serological testing for ILD will ultimately save unnecessary expenses associated with general laboratory screening. Finally a broader consensus on the strategy for laboratory testing in ILD in general might also improve the detection level of these relatively rare diseases and this will ultimately improve
management and care of patients suffering from these complex disorders.

Acknowledgment

TB, MR, FG, MC and KFR performed the literature search, designed the figures, contributed to the manuscript, and approved its final version.

Glossary

Abbreviations and eponyms

<table>
<thead>
<tr>
<th>Term</th>
<th>Definition</th>
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<tr>
<td>AAV</td>
<td>ANCA-Associated Vasculitis</td>
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<td>ASS</td>
<td>Anti-Synthetase Syndrome</td>
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<td>C3/C4</td>
<td>Complement protein 3/4</td>
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<td>CK</td>
<td>Creatine Kinase</td>
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<td>CTD</td>
<td>Connective Tissue Disease</td>
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<td>CREST</td>
<td>Calcinosis, Raynaud’s phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia</td>
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<td>DPLD</td>
<td>Diffuse Parenchymal Lung Disease</td>
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<td>ELISA</td>
<td>Enzyme Linked Immuno Assay</td>
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<td>EIA</td>
<td>Enzyme Immune Assay</td>
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<td>ENA</td>
<td>Extractable Nuclear Antigens</td>
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<td>ILD</td>
<td>Interstitial Lung Disease</td>
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<td>IIF</td>
<td>Indirect Immunofluorescence</td>
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<td>LDH</td>
<td>Lactate Dehydrogenase</td>
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<td>LIA</td>
<td>Line Immunoassay</td>
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<td>MCTD</td>
<td>Mixed Connective Tissue Disease</td>
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<td>MPA</td>
<td>Microscopic Polyangiitis</td>
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<td>SLE</td>
<td>Systemic Lupus Erythematoses</td>
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<td>SSc</td>
<td>Systemic Sclerosis</td>
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<td>dcSSc</td>
<td>diffuse cutaneous Systemic Sclerosis</td>
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<tr>
<td>lcSSc</td>
<td>limited cutaneous Systemic Sclerosis</td>
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<tr>
<td>UCTD</td>
<td>Undifferentiated Connective Tissue Disease</td>
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<td>U, RNP</td>
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<td>Sm</td>
<td>Sm</td>
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<td>ds-DNA</td>
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<td>Topo-I</td>
<td>Topo-I</td>
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<tr>
<td>Th/To, CENP-B RNA Polymerase III</td>
<td>Th/To, CENP-B RNA Polymerase III</td>
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<td>PL 7, PL 12</td>
<td>PL 7, PL 12</td>
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<tr>
<td>OJ, EJ, KS, Mi-2, MDA-5</td>
<td>OJ, EJ, KS, Mi-2, MDA-5</td>
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<td>PM/Scl-100</td>
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**Fig. 1.** Suggested diagnostic algorithm for the serological approach to the patient presenting with unclear ILD. In the presence of any suspicious clinical signs or symptoms, reflective of an underlying CTD, early consultation of an experienced rheumatologist is recommended, even in the absence of any positive laboratory results. ASS: Anti-synthetase syndrome. CREST (syndrome): Calcinosis, Raynaud’s phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia. MCTD: Mixed Connective Tissue Disease. MPA: Microscopic Polyangiitis. RA: Rheumatoid Arthritis. SLE: Systemic Lupus Erythematoses.
Abiotypes and Antibody-Tests

ANA
Anti-Nuclear Antibodies

ANCA
Anti-Neutrophil Cytoplasmic Antibodies

MPO-ANCA
Myeloperoxidase Anti-Neutrophil Cytoplasmic Antibodies

CADM-140
Anti - Clinically Amyopathic DM polypeptide Antibody (140 kd) aka. MDA-5

CENP-B/F
Centromere Protein B/F antibody

CCP
Cyclic Citrullinated Peptide antibody
dsDNA
double strand Deoxyribonucleoprotein antibody

EJ
Glycol antibody

Jo-1
Histidyl antibody

KS
Asparaginyl antibody

Mi-2
Nucleosome remodeling-deacetylase (NuRD) complex — antibody

NuMA
Nuclear matrix protein

OJ
Isolecyl antibody

PL-7
Threonyl antibody

PL-12
Alanyl antibody

PM/ScI-75/100
Polyomvits-citrooler disorder antibody (75/100 kDa), human exosome complex

RF
Rheumatoid Factor

RNA-Polymerase III
n/a

SSA
Sjogren Syndrome-A antibody (Ro ribonucleoprotein: Ro52/Ro60)

SSB
Sjogren Syndrome-B antibody (La ribonucleoprotein domain family)

Sm
Smith antibody (Small Nuclear Ribonucl eoprotein Core Proteins)

Th/To
Proteins of the RNase MRP and RNase P Ribonucleoprotein Complex

Topo-I
Topoisomerase I (aka. SCL-70: scleroderma 70 antibody)

U1 RNP
U1-Ribonucleoprotein antibody

Conflict of interest

The authors declare that they have no competing interests.

References


Y. Itoh, M. Reichlin, Autoantibodies to the Ro/SSA antigen are conformation dependent: I. Anti-60 kDa antibodies are mainly directed to the native protein; anti-52 kDa antibodies are mainly directed to the denatured protein, Autoimmun. Rev. 14 (1) (1992) 57–65.


L. Weinirb, O.P. Sharma, F.P. Qasimroo, J.A. Long-term study of interstitial lung disease in systemic lupus erythematosus, Semin. Arthritis Rheum. 20...