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CONTEMPORARY REVIEW

Diagnostic Approach for Suspected Acute Myocarditis: Considerations for Standardization and Broadening Clinical Spectrum

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ABSTRACT: Myocarditis is most recognized in patients with moderate to severe, recent-onset heart failure. However, less typical presentations including myocardial infarction with normal coronary arteries and arrhythmias are important manifestations but less commonly recognized to be caused by myocarditis. Most cases of myocarditis can be self-limiting without specific treatment; however, appropriate identification of risk during the diagnostic process of myocarditis and once a diagnosis is established is of primordial importance to identify patients in need for more specific follow-up and management. We propose a flexible, multitiered approach to the diagnostic process, allowing for capturing of the spectrum of myocarditis at an early time-point, individualized use of diagnostic resources through disease severity phenotyping, and providing structured follow-up care once myocarditis is confirmed. Such diagnostic processes allow for identification of specific etiologies with potential therapeutic consequences or allows for the comprehension of disease chronicity by understanding genetic contributions or elements of persistent immune dysregulation and degree of cardiac damage. The article highlights the evolving field of immunophenotyping in myocarditis, generating a potential for the development of targeted therapeutic approaches. Currently long-term follow-up should be titrated to the refined risk assessments of patients with a diagnosis of myocarditis and includes arrhythmia monitoring and imaging when the results will likely impact management. Genetic testing should be considered in selected cases, and histologic diagnosis may be considered in nonresponders even at later stages.

Key Words: cardioimmunology ■ care pathway ■ genetics ■ management ■ myocarditis

Myocarditis remains a challenging diagnosis because of the heterogeneity in the clinical manifestation and severity of the disease. Several common scenarios as myocardial infarction with normal coronary arteries and frequent premature ventricular complexes (PVCs) do not always trigger a search for underlying causes, and as a result, the true incidence of myocarditis remains uncertain. This review aims to provide a critical appraisal on the broader clinical scenarios in which myocarditis may be plausible and addresses the evolving approaches in diagnosing myocarditis, offering a standardized patient-centric approach using tiered evaluation. We first highlight some of the current challenges in the field of diagnosing myocarditis and secondly illustrated how a standardized approach can circumvent potential challenges. The therapeutic management of myocarditis falls beyond the scope of this article and has been reviewed in more depth for specific causes of myocarditis elsewhere.[1,2]

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Nonstandard Abbreviations and Acronyms

EMB	endomyocardial biopsy
LGE	late gadolinium enhancement
P/LP	pathogenic/likely pathogenic

CURRENT CHALLENGES IN MYOCARDITIS

Challenges in Defining Myocarditis

Myocarditis is an inflammatory disease of the heart that can be provoked by a wide variety of infectious and noninfectious triggers (Figure 1; Table S1), leading to an immune-mediated inflammatory reaction of the heart in a susceptible host.[3] A definitive diagnosis of myocarditis is made when a patient presents with a clinical context compatible with myocarditis and cardiac inflammation either on endomyocardial biopsy (EMB) or cardiac magnetic resonance (CMR) imaging is present (Figure 2). When such inflammation persists over time, a scar may form, increasing the risk for malignant arrhythmias and cardiac dysfunction, generating the basis of chronic inflammatory cardiomyopathy. The transition into a chronic inflammatory cardiomyopathy can occur in up to 30% of biopsy proven myocarditis.[4]

Several distinct and potentially modifiable pathways link acute myocarditis to the common phenotype of a chronic cardiomyopathy. But in general when left ventricle (LV) dysfunction develops the patient either develops persistent inflammation (immune polarization in Figure 1)[5,6] and/or harbors pathogenic/likely pathogenic (P/LP)-variants in desmosomal or other dilated cardiomyopathy genes forming a multi-hit to the myocardium.[7,8] A more detailed description on the process of immune tolerance, immune polarization and role of immunophenotyping is provided in Data S1. Such revised understanding of the pathophysiology of myocarditis helps to understand further diagnostic approaches in defining myocarditis and highlight potential evolutions in the management of myocarditis based upon immunophenotyping (Figure 1).

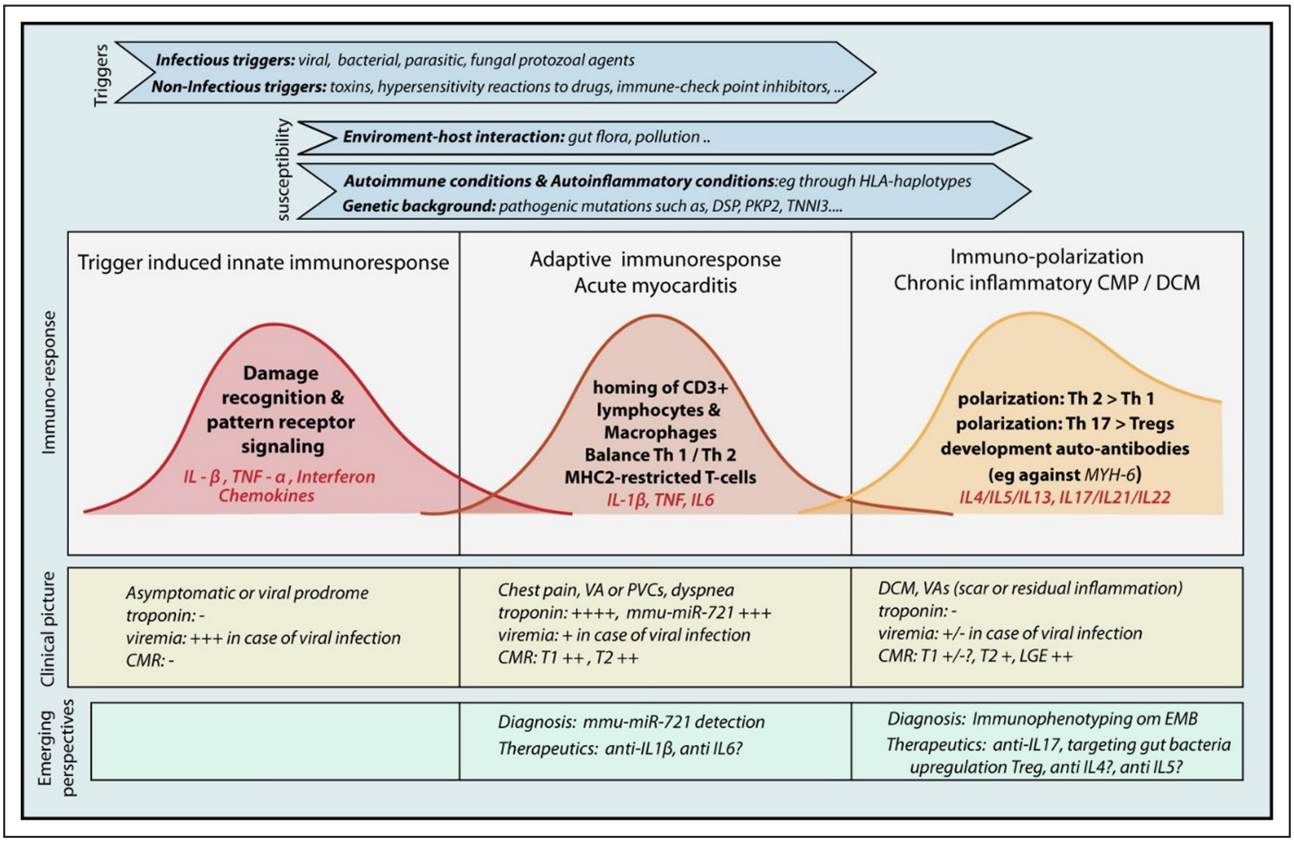

Figure 1. Immunophenotypic background of myocarditis.
Triggers induce an innate and adaptive immune response, while susceptibility factors such as host-environment interaction and genetic background contribute to the severity and chronicity of this event. The clinical picture of acute myocarditis often coincides with the adaptive immune response. DCM indicates dilated cardiomyopathy; *DSP*, desmoplakin; *HLA*, human leukocyte antigen; IL, interleukin; LGE, late gadolinium enhancement; MHC, major histocompatibility complex; Mmu-miR, *Mus musculus* microRNA; MYH 6, myosin heavy chain, α isoform; *PKP2*, plakophilin-2; PVC, premature ventricular complexes; Th, T-helper lymphocyte; *TINNI3*, cardiac troponin I; TNF, tumor necrosis factor; and VA, ventricular arrhythmias.

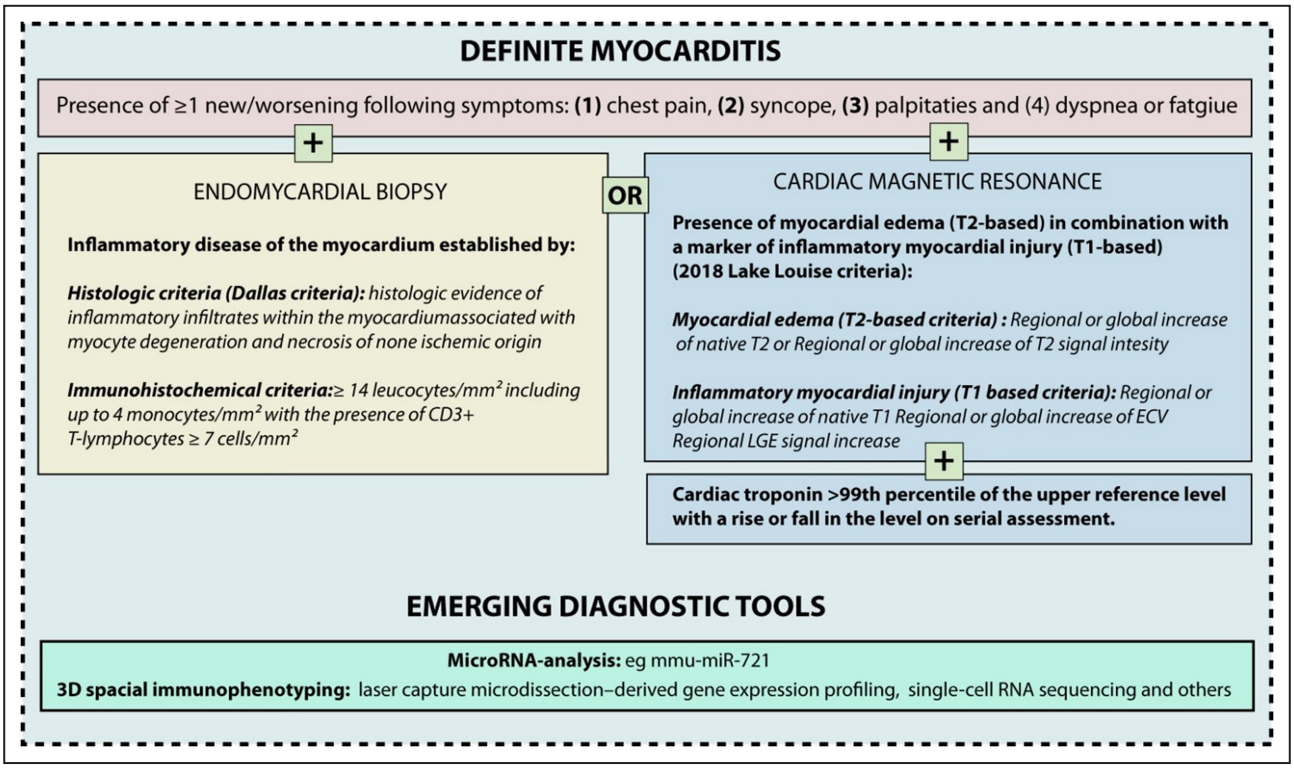

Figure 2. Definition of definite myocarditis.
ECV indicates extracellular volume; and RNA, ribonucleic acid.

Challenges in Capturing the Full Clinical Spectrum of Myocarditis

While definite acute myocarditis forms the tip of the iceberg of the myocarditis spectrum, a large proportion of patients in clinical practice do not undergo myocarditis testing, leading to missed opportunities in diagnosis (Figure 3).[9] The wide spectrum of clinical manifestations of myocarditis (chest pain, dyspnea/heart failure, syncope or arrhythmia-related) results that patients will be seen across multiple cardiology subspecialties. Raising clinical suspicion of myocarditis is essential to establish a diagnosis of definite myocarditis.

For example, around 33% of patients with myocardial infarction with normal coronary arteries may get a diagnosis of myocarditis based on CMR assessment.[10] Similarly, myocarditis can be found in patients with frequent PVCs or ventricular arrhythmias (VAs).[11,12] Detailed analysis with CMR or fluroredeoxyglucose positron emission tomography (FDG-PET) in patients with frequent PVCs (not attributable to other reasons) illustrates a diagnosis of myocarditis in up to 38% to 51%.[11,12] Myocarditis has been found in around 10% of patients with a nonischemic cardiomyopathy undergoing VA ablation.[13] Similarly, third degree atrioventricular block in younger patients (<55 years) should trigger a search for specific forms of myocarditis (sarcoidosis, giant cell myocarditis, Lyme carditis).[3] Finally, EMB analysis in patients with a chronic (>3 months) unexplained cardiomyopathy reveals underlying myocarditis in 9% to 40% of patients.[14]

Based on this broad clinical spectrum, we propose a standardized referral and diagnostic process towards myocarditis (Figure 3). Establishing a diagnosis of definite myocarditis has numerous diagnostic and therapeutic implications, including (1) temporary prohibition of competitive sports, (2) a search for specific forms of myocarditis that need targeted therapies (eg, immunosuppressive therapies in sarcoidosis, eosinophilic, giant cell myocarditis, and immune checkpoint inhibitors or antibiotics in Lyme carditis), (3) timing of diagnostic tests such as CMR before device implantation, and (4) genetic screening, counseling, and potential cascade screening.

Challenges in Classifying Myocarditis

Several nonmutually exclusive approaches in clinical practice are used to classify myocarditis. This includes clinicopathological criteria (Lieberman criteria),[15] histopathologic criteria (such as the Dallas criteria),[16] classification based on the type of inflammatory infiltrate (Table 1), classification based on the presence of cardiac viral infiltrate (viral-positive or viral-negative) and the tropism of the virus (Table 1), and classification based on the duration of symptoms (acute or chronic myocarditis) or the severity of the presentation (fulminant vs nonfulminant myocarditis).[17,18] A clinically

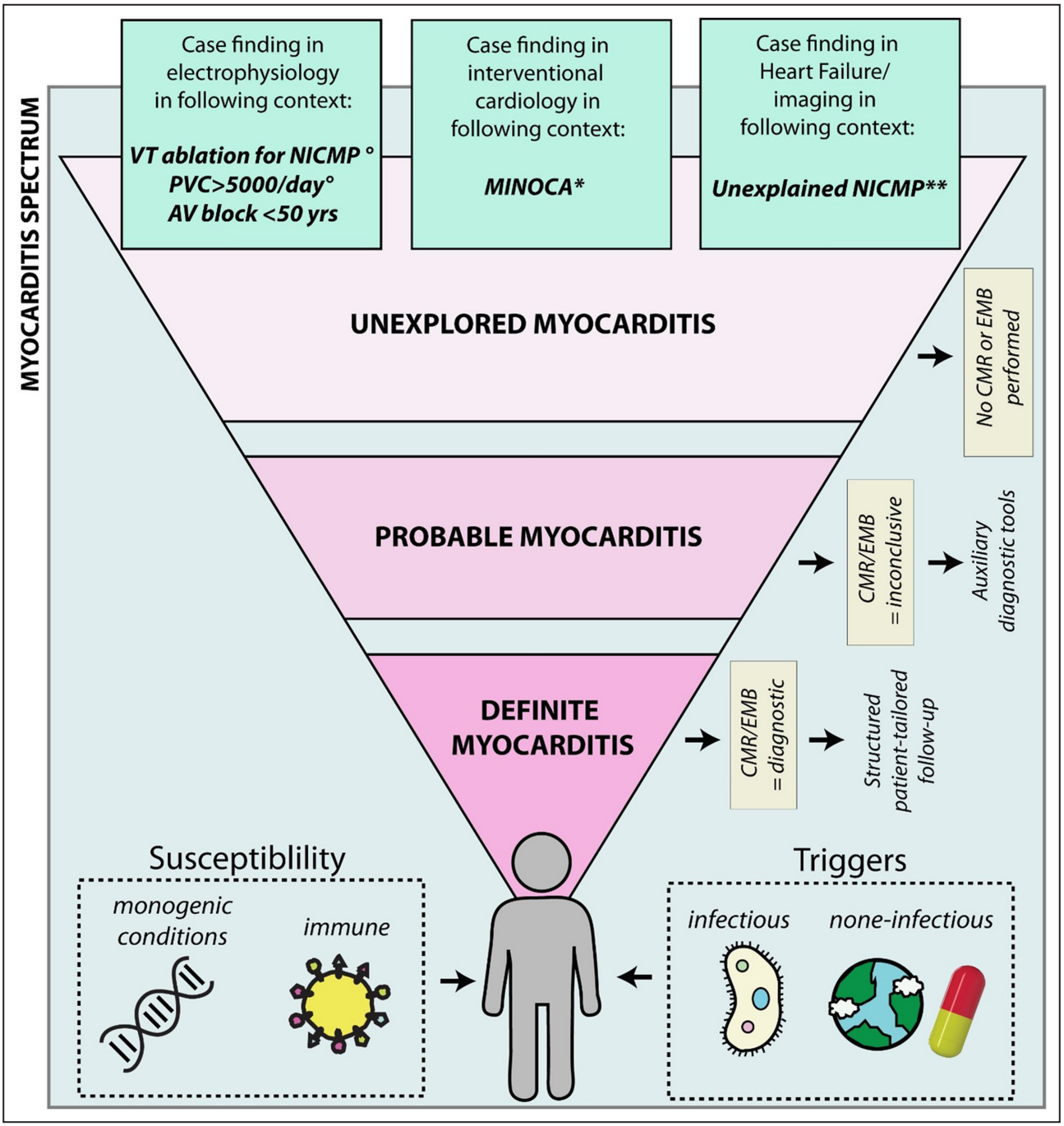

Figure 3. Clinical spectrum of myocarditis.
Probable myocarditis is the setting in which endomyocardial biopsy or cardiac magnetic resonance imaging generates borderline/inconclusive results. Definite myocarditis includes cases with a confirmed diagnosis in line with Figure 2. Unexplored myocarditis includes cases in which no confirmatory investigations (endomyocardial biopsy or cardiac magnetic resonance imaging) regarding the presence of myocarditis are performed. CMR indicates cardiac magnetic resonance imaging; EMB, endomyocardial biopsy; MINOCA, myocardial infarction with nonobstructive coronary arteries; NICMP, nonischemic cardiomyopathy; PVC, premature ventricular complexes; VT, ventricular tachycardia; °, case finding in electrophysiology; *, case finding in interventional cardiology; and **, case finding in heart failure and imaging.

useful approach for classification of myocarditis should center the patient, integrating available knowledge on their risk and generate a framework for individualized management.

A classification based on the initial presentation has been shown to meaningfully predict prognosis. Large multicenter registries show that patients who present with fulminant myocarditis (cardiogenic shock) or

Table 1. Classification of Myocarditis According to the EMB Result of Inflammatory Cell Infiltrate or Tropism of the Viral Infiltrate

Type	Histologic features	Main manifestation forms	EMB comments	Advanced imaging*	Therapeutic implications
Classification according to inflammatory infiltrate					
Giant cell myocarditis	Necrotizing inflammation with multinucleated giant cells and lymphocytes, eosinophils and histiocytes. Absence of granulomas.	Acute and fulminant presentation with cardiogenic shock, VT and possible third-degree atrioventricular block. Progressive over short period of time requiring MCS.	EMB has high diagnostic yield attributable to large area of affected myocardium (±80%).	Limited role of advanced imaging as presentation is common cariogenic shock. Limited data with MRI suggest widespread LGE affecting all myocardial layers.	- Multidrug immunosuppression upfront - inotropes and MCS in case of cardiogenic shock. - Consider urgent transplant evaluation. - ICDs considerations
Cardiac sarcoidosis	Granulomas in the absence of infection. Eosinophils and necrosis are normally absent.	Third-degree atrioventricular block, VA, cardiomyopathy (mostly moderate reduced LVEF), supraventricular arrhythmias.	EMB sensitivity can be increased using EVM. Region with low fractionated voltage often coincide with regions of LGE.	CMR: patchy LGE intramyocardial and sub-epicardial involvement, primarily of the basal septum and inferolateral wall of the LV.	- Immunosuppression: first-line steroids, if FDG-PET uptake persists: +MTX - CIED according to specific recommendations - If LVEF is low, classic guideline recommended GDMT
Eosinophilic myocarditis	Endocardial and perivascular forms. Forms with extensive myocytes damage (necrotizing eosinophilic myocarditis).	Heart failure attributablee to restrictive cardiomyopathy or valve involvement. Embolization of thrombi is common.	Consider biventricular biopsy as disease might be predominantly affecting LV apex, with reduced sensitivity of RV biopsy.	CMR documents myocardial infiltration with LGE and edema with T2. Often involvement mitral leaflets and papillary muscle. CMR visualizes endomyocardial involvement.	- Corticoid steroids - Drug discontinuation in case of allergic reaction - MCS if shock - Specific forms might need specific therapies†
Lymphocytic myocarditis	Lymphocyte predominant, nonischemic infiltrate with/or without myocyte necrosis (borderline if no myocyte damage).	Fulminant cases present with shock and nondilated LV. Nonfulminant cases most often have dilated LV and heart failure.	EMB often performed classically in RV. Limited data on EVM or imaging targeted approach.	Classic Lake Louise CMR findings.	Therapy is predominantly supportive (inotropes MCS if shock or GDMT if heart failure) with no administration of immunosuppressive therapy.
Classification of viral-positive myocarditis according to the tropism to the heart					
Viral tropism mechanism	Cardiotropic	Vasculotropic	Lymphotropic	Cardiotoxic	ACE2-tropic
Examples	Adenovirus, enteroviruses	Parvo B19	CMV, EBV, HVV6	HCV, HIV, influenza	MERS-CoV, SARS-CoV, SARS-CoV-2

ACE2 indicates angiotensin-converting enzyme 2; CIED, cardiovascular implantable electronic device; CMR, cardiac magnetic resonance; EMB, endomyocardial biopsy; EVM, endocardial voltage mapping; FDG-PET, fluroredeoxyglucose positron emission tomography; GDMT, guideline-directed medical therapy; HCV, Hepatitis C virus; ICD, implantable cardioverter-defibrillator; LGE, late gadolinium enhancement; LV, left ventricle; LVEF, left ventricular ejection fraction; MCS, mechanical circulatory support; MRI, magnetic resonance imaging; MTX, methotrexate; RV, right ventricle; VA, ventricular arrhythmia; and VT, ventricular tachycardia.

*Beyond echocardiography.

†For example, cyclophosphamide in eosinophilic granulomatosis with polyangiitis-related eosinophilic myocarditis, imatinib in myeloproliferative variant.

complicated nonfulminant myocarditis (sustained ventricular arrhythmia, atrioventricular block, or LV dysfunction with an LVEF <50%) exhibit a rate of cardiac mortality or heart transplantation of 10.4% at 30 days and 14.7% at 5 year follow-up.[19] While patients with nonfulminant/uncomplicated acute myocarditis had no cardiac mortality or heart transplant at follow-up.[19] This underscores that the adverse risk associated with myocarditis is predominantly early and dictated by the severity of the initial manifestation.

For patients presenting with complicated/fulminant myocarditis the underlying type of inflammatory infiltrate further defines prognosis and identifies patients for specific immunosuppressive therapies (Table 1).[4] An emerging concept in the classification of myocarditis is the identification of specific immunophenotypes and inflammatory pathways in the process of developing myocarditis and transitioning from acute myocarditis to a chronic inflammatory cardiomyopathy (immune polarization) as illustrated in Figure 1.[6,20] For instance, polarization towards Th-17 immune response occurs in acute myocarditis[6] and more recently it has been shown that detection of microRNAs (mmu-miR-721) produced by this subset of cells can differentiate from myocardial infarction with great diagnostic accuracy (C-statistic=0.927).[21] Such emerging concepts in classification could have an important impact on the diagnosis, classification and selection of potential treatments in myocarditis as reflected in Figure 1.

STREAMLINED DIAGNOSTIC APPROACH TO MYOCARDITIS

The diagnostic approach in any care pathway for myocarditis needs to be individualized to the clinical setting in which patients present (outpatient versus inpatient) and titrated to the disease severity, as the latter as a major determinant of immediate risk. Herein, we propose a 3-tier diagnostic approach consisting of: (1) a first tier of investigations to be performed in all patients, aimed at understanding the immediate prognosis; (2) a second tier of investigations aimed at confirming the diagnosis of definite myocarditis; and (3) a third tier of auxiliary diagnostic evaluations and titration of care based on the assessment during the first 2 tiers.

Tier 1: Acute Evaluation

In all patients presenting with suspected myocarditis a detailed history taking, physical evaluation, laboratory analysis, ECG, and echocardiography should be done (Figure 4).[14] None of these elements have sufficient sensitivity and specificity to establish a diagnosis of myocarditis; however, they are important in excluding other causes of the acute presentation. Such evaluation also allows for risk assessment for patients presenting as inpatient or outpatients. Essential elements of this tier of evaluation are listed in Table 2. Analysis of different cohorts of patients with myocarditis indicate that chest pain is a presenting complaint in 27% to 89% of patients, dyspnea/heart failure/LV-dysfunction in 19% to 80% and arrhythmias or syncope in 6% to 22%.[4,19,22,23] Up to 80% of patients have a prodromal episode (eg, gastrointestinal symptoms, sore throat) before admission.[3] A history of myocarditis is infrequent (around 1%, but maybe more common in settings of repetitive toxic exposure such as cocaine use). A history of autoimmune or autoinflammatory conditions is relatively frequent (Table 2).[4,17,19] Table S2 lists clinical pearls of most common autoimmune or autoinflammatory conditions. Myocarditis associated with drug or vaccine administrations usually occurs within a few days after exposure. A detailed description of COVID-19 myocarditis or COVID-19 vaccine–related myocarditis falls beyond the scope of this article but has been provided more recently.[24]

Electrocardiographic analysis is usually abnormal, showing diffuse ST-segment elevations (classically concave without reciprocal changes) or T-wave abnormalities, occurring in more than half of patients.[3] PVCs occur in around 25% and especially polymorphic PVCs raise a possibility to myocarditis.[12] Conduction abnormalities such as atrioventricular block might hint towards specific forms of myocarditis (eg, sarcoidosis, Lyme carditis). Patients with fulminant myocarditis more often manifest with a left bundle branch block. A QRS prolongation above 120 ms or presence of Q-waves are associated with adverse prognosis.[19] A normal electrocardiographic does not exclude myocarditis and can occurring in up to 15%, especially in patients with uncomplicated myocarditis. Laboratory markers including inflammatory markers (C-reactive protein or erythrocyte sedimentation rate), cardiac troponins, and natriuretic peptides are often elevated, but normal values do not exclude myocarditis and when elevated are not specific.[25] Viral titers are often ordered, but elevated viral titers do no confirm or refute specific viral causes of myocarditis. Elevated admission and peak troponins (plasma cardiac troponin T >50 ng/L) are associated with worse prognosis, but only show moderate correlation with admission LVEF.[26] Echocardiography is useful in excluding other causes of the presenting symptoms. Echocardiography offers quick bedside evaluation of LVEF, global longitudinal strain, regional wall motion abnormalities, presence of pericardial fluid, the degree of LV dilatation, hypertrophy, and diastolic dysfunction. Presence of a reduced admission LVEF <50% offer prognostic information in patients with myocarditis allowing for initial triaging (Figure 4).[19] In general patients with suspected myocarditis need to be admitted, with patients presenting with fulminant and complicated myocarditis requiring intensive care unit monitoring.

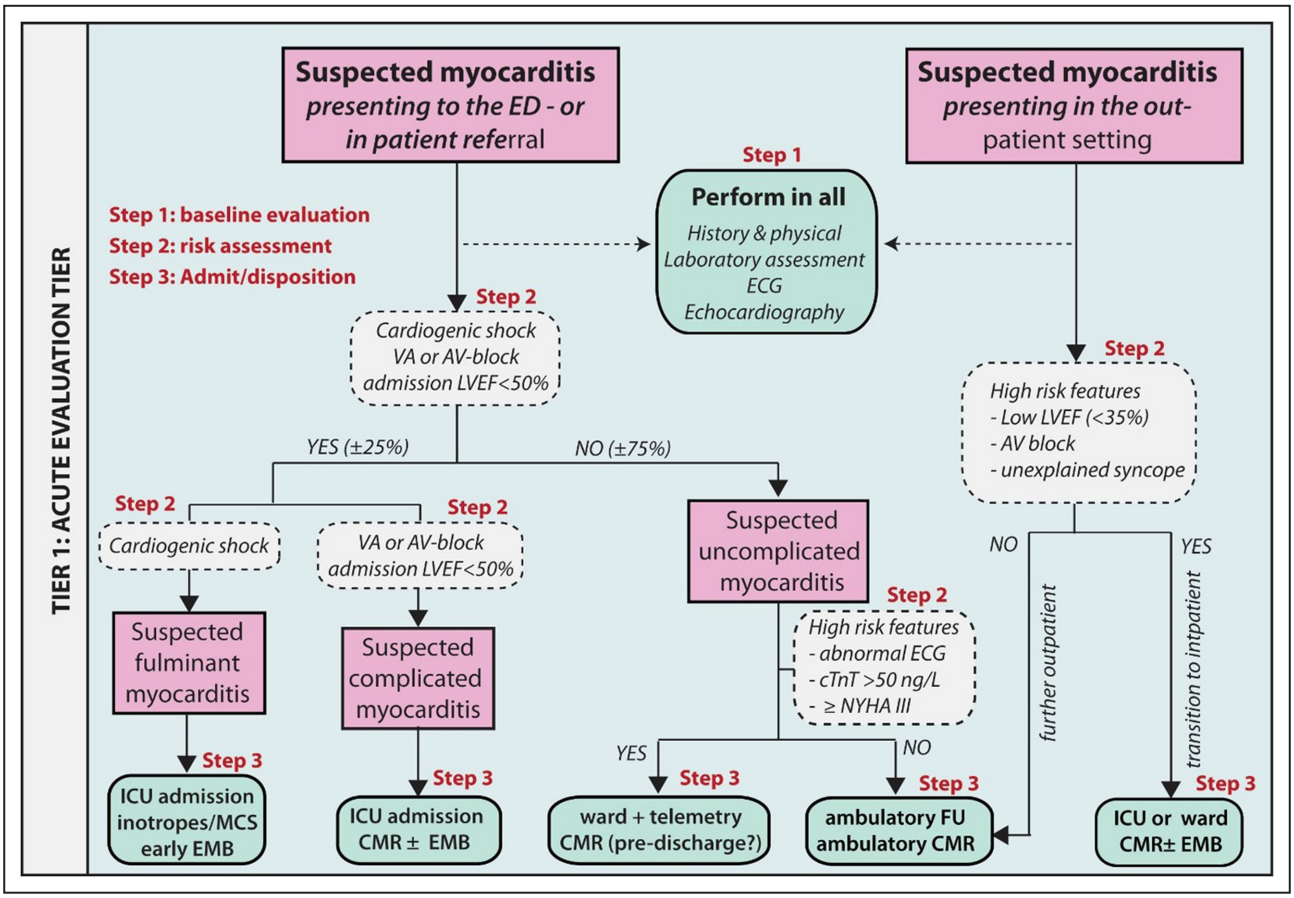

Figure 4. Tier 1—acute diagnostic evaluation.
AV indicates atrioventricular; CMR, cardiac magnetic resonance imaging; cTnT, cardiac troponin T; ED, emergency department; EMB, endomyocardial biopsy; EVM, endomyocardial voltage mapping; ICU, intensive care unit; LV, left ventricle; LVEF, left ventricular ejection fraction; MCS, mechanical circulatory support; NYHA, New York Heart Association Class; and VA, ventricular arrhythmia.

Patients with uncomplicated myocarditis with higher risk features (Figure 4) need to be admitted to units with telemetry capability, given the risk of ventricular arrhythmias or atrioventricular conduction abnormalities. Patients with uncomplicated myocarditis without high-risk features can potentially be managed on an ambulatory basis.

Tier 2: Confirmation of Myocarditis

After a first-tier evaluation that allows the understanding of the immediate prognosis, a second tier of evaluation consists of establishing the diagnosis of definite myocarditis (Figure 5). The common finding of elevated troponin, ST-segment and T-wave abnormalities, chest pain, and regional wall motion abnormalities, renders coronary artery disease a frequent differential diagnosis, often necessitating a coronary angiogram for its exclusion. The younger age of patients with suspected myocarditis (mean age, 34–47 years in large series), argues for individualization of the diagnostic approaches to exclude coronary artery disease. The emerging understanding of the role of Th17 in myocardial injury in myocarditis, but not in myocardial infarction could become of clinical utility in the future as a recently identified microRNAs (mmu-miR-721) produced by the Th17 subset differentiate with great diagnostic accuracy between both conditions.[21] As outlined in Figures 2 and 5, in patients with suspected myocarditis a confirmation of the diagnosis needs to be attained using either EMB or CMR.

Endomyocardial Biopsy

EMB remains the gold standard diagnostic approach to determine the etiology of the cardiac inflammation. A recent joint position statement described scenarios in which characterization of the underlying etiology of the inflammation results in important therapeutic consequences hereby justifying the performance of an EMB.[27] For myocarditis specifically, tier 1 based risk evaluation dictates the urgency of performing an EMB (Figure 5). The specific inflammatory infiltrate found on EMB (Table 1) also confers prognostic information, with

Table 2. Essential Checklist of Tier 1 Diagnostic Evaluations

Question	Explanation
History and physical examination	
History of cancer	Especially use of immune checkpoint inhibitor and anthracyclines.
History of asthma	Up to 17% of patients can have an allergic background.
History of autoimmune disease or autoinflammatory conditions	Around 7%–9% of patients have a history of autoimmune disorder. Autoinflammatory conditions are also associated with myocarditis/pericarditis.
Family history of cardiomyopathy	Unclear of % myocarditis patients have family history of cardiomyopathy; however, common genetic backgrounds are found (eg, desmoplakin-mutation). Establishing pedigree is important for potential cascade screening.
Chief complaint	Chest pain, dyspnea, and arrhythmia/syncope. Type of complaint infers little over short-term prognosis, however ≥NYHA 3 is associated with adverse short-term prognosis.
Medication history	Exposure to medications associated with myocarditis: cephalosporins, digoxin, diuretics, dobutamine, sulfonamides, tricyclic anti-depressants.
COVID-19 and vaccination	Recent SARS-CoV2 infection or mRNA vaccination for SARS-CoV2? Chicken pox vaccine or others.
Abuse and toxins	Cocaine and amphetamine use, ethanol consumption, exposure to copper, iron, lead, venomous bites.
Test	**Explanation**
Laboratory analysis	
White blood cell count+distribution	Inflammatory marker often elevated; peripheral eosinophils often elevated in eosinophilic myocarditis (normal does not exclude eosinophilic myocarditis).
C-reactive protein	Elevated in 80%–95% of patients.
Myocardial necrosis markers	High-sensitive troponins are often elevated; plasma cTnT >50 ng/L identifies patients with worse prognosis. CK-MB often elevated. CK-MM fraction is also useful to detect potentially associated myositis (can complicated diagnostic accuracy of CMR, which uses skeletal muscle as reference).
Erythrocyte sedimentation rate	Often elevated. If persistently elevated might hint towards underlying autoimmune disorder.
Biomarkers	NT-proBNP offers prognostic information and is often elevated. ST2 offers prognostic and mechanistic insights predominantly in male patients.
Autoimmune screening	Should be considered in specific cases, and the individual case determines the specific choice but can potentially include ANA, anti-dsDNA, cardiolipin antibody IgG/IgM, 1, 25 dihydroxy vitamin D, ANCA/P-ANCA, myeloperoxidase, PR-3 antibody, parathyroid hormone, soluble interleukin-2 receptor, rheumatoid factor, ACPA, SPEP, anti-CCP, anticentromere, anti-Scl-70, anti-RNA polymerase III, anti-SSA, anti-SSB, specific inflammatory myositis antibodies.
Viral serology	Positive viral serology does not indicate myocardial infection. Viral serology screening is not recommended in all patients. In selected cases serology can be useful, such as HCV, HIV, and Lyme serology and influenza testing, as these would require direct treatment.

ACPA indicates anti-citrullinated protein antibody; ANA, antinuclear anibody; ANCA, antineutrophil cytoplasmic antibodies; anti-CCP, anti–citrullinated protein antibody; anti-SSa, anti-Ro; anti-SSb, anti-La; CK-MB, creatinine kinase muscle brain; CK-MB, creatinine kinase muscle muscle; CMR, cardiac magnetic resonance imaging; cTnT, cardiac troponin T; dsDNA, double-stranded DNA; HCV, hepatitis C virus; IgG, immunoglobulin G; IgM, immunoglobulin M; NYHA, New York Heart Association Class; NTproBNP, aminoterminal pro-B-type natriuretic peptide; PR3, anti-proteinase-3; SPEP, serum protein electrophoresis; and ST2, soluble interleukin 1 receptor-like 1.

graded decrease in severity from giant cell myocarditis, to eosinophilic myocarditis, to cardiac sarcoidosis and to lymphocytic myocarditis.[17] The independent prognostic role of presence of viral genome beyond immunologic evidence of inflammation has not been consistently reproduced and anti-viral therapy has yet to provide consistently incremental benefit. Therefore, discussion exists about the role of detection of viral genome on EMB analysis as part of routine care. But finding of cardiotoxic viruses (Table 1) might have therapeutic implications. Best practices on performing EMBs are reflected in Figure 5. Taking these elements into account, a recent meta-analysis (10 491 patients) indicated a detection rate of inflammation in EMB specimens in around ~51%.[28] In experienced centers, EMB can be performed safely with major complication rates <1% to 2%. However at lower volume centers complication rates up to 8.9% are reported.[27] Despite safety of EMB in experienced centers, the rate of EMB has declined over the years indicating that EMBs are performed only in 3% of all suspected acute myocarditis cases.[29] A trend which is partially explained by the wide availability and noninvasive nature of CMR. With advancements in the field of myocardial tissue analysis such as 3D-spatial immunophenotyping using laser capture microdissection derived gene expression profiling and single cell RNA sequencing, EMBs may someday be an essential tool for individualization of treatment approaches (Figure 2).

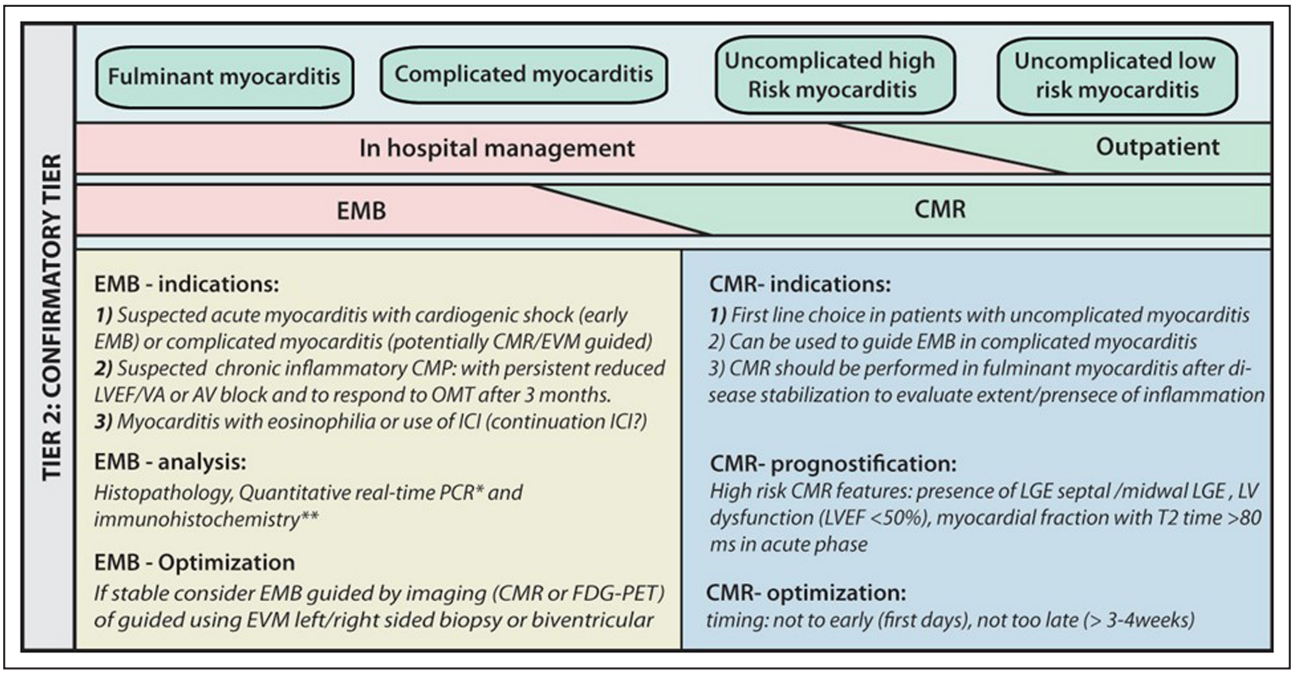

Figure 5. Tier 2—confirmation of myocarditis.
CMR indicates cardiac magnetic resonance imaging, mapping; EMB, endomyocardial biopsy; EVM, endocardial voltage mapping; FDG-PET, fluroredeoxyglucose positron emission tomography; ICI, immune checkpoint inhibitors; LGE, late gadolinium enhancement; LV, left ventricular; OMT, optimal medical therapy; PCR, polymerase chain reaction; *, case finding in interventional cardiology; and **, case finding in heart failure and imaging.

Cardiac Magnetic Resonance Imaging

CMR is the current gold standard noninvasive method used for the diagnosis of myocarditis. Unless formal contraindications exist, all patients with myocarditis should undergo a CMR scan (even patients with EMB-proven myocarditis). Nevertheless, in clinical practice waiting times for CMR and regional access to CMR do form barriers to such universal implementation. Figure 5 gives an overview of the indications for CMR in the setting of acute myocarditis. CMR provides strong evidence for myocarditis if myocardial edema is documented (T2-based evaluation) combined with a marker of inflammatory myocardial injury (T1-based).[30] As such it is important that both T1 and T2 sequences are adequately performed, which might not always be performed in clinical practice. In comparison to the original 2009 Lake Louise criteria, the updated 2018 criteria (see Figure 2) show increased sensitivity (87.5% versus 74% previously) together with high specificity for acute myocarditis (96.2% versus 86% previously).[30] However, the diagnostic accuracy also depends on the clinical presentation form of the patient, with highest sensitivity in myocardial infarction with normal coronary arteries manifestation and lowest in arrhythmia presentation (Figure 5).[31] Myocardial edema also diminishes over time and therefore the highest diagnostic performance of CMR is within 2 to 3 weeks after the onset of symptoms. A normal extracellular volume in the acute setting is the most sensitive marker in excluding myocarditis (AUC=86.3%).[32] Tissue characterization with CMR might also be less sensitive in the very acute phase (first days) or when there is associated myositis (as normal skeletal muscle is used as a reference). In comparison to EMB, CMR cannot determine the type of inflammatory infiltrate or detect the presence of a viral infiltrate.

CMR additionally offers prognostic information by giving insights into presence of LGE, the location of LGE (with mid-wall LGE conferring a worse prognosis), presence of LVEF <50% and acute phase myocardial fraction with T2-time >80 ms.[25,33] LGE occurs in around 36% to 44% of patients with myocarditis and offers incremental prognostic information even when LVEF is normal.[25] Next to tissue characterization and defining prognosis, CMR can also assess the presence of specific cardiomyopathies (eg, cardiac sarcoidosis; Table 1) or exclude cardiac ischemia. However, little data are available about the role of perfusion CMR in helping to exclude the presence of cardiac ischemia during myocarditis evaluation.

Challenge of Outpatient Assessment

Both CMR and EMB evaluations within 2 weeks of onset of symptoms have the highest diagnostic acuity (coinciding with peak inflammation and myocardial injury as reflected in Figure 1). As patients in the

outpatient setting often have a longer duration of symptoms, classic myocarditis features on CMR (or EMB if performed) can be absent (probable myocarditis in Figure 3). Potentially other techniques such as FDG-PET could be useful to detect persisting inflammation. In the outpatient setting, searching for factors associated with chronicity of the disease such as search for specific autoimmune conditions (associated with persistent inflammation) and predisposing P/LP variants, is often important in understanding the entire picture. In patients with a semi-recent cardiomyopathy unresponsive to standard medical therapy (>3 months), guidelines suggest performing an EMB, which might detect the presence of ongoing inflammation.

Tier 3: Discharge and Follow-Up

Patients are classically discharged when the clinical situations are stable (hemodynamic stability, controlled ventricular arrhythmia and atrioventricular conduction abnormalities) and cardiac troponin values are normalizing. It is advised against exercise testing predischarge, as exercise can elicit VAs in the acute setting.[14] The intensity of follow-up in patients with established myocarditis is determined by the presence of low- and high-risk features (Figure 6). Acute myocarditis resolves completely in 2 to 4 weeks in over 50% of patients, especially in the presence of low-risk features. However, in settings with high-risk features around 25% will develop persisting LV dysfunction. Five-year combined events rate of mortality and transplant are up to 14.5% in complicated myocarditis and 43% in fulminant myocarditis, driven by, respectively, 5% and 14.5% transplant rate.[17,19] The initial presentation has been shown to confer prognosis with heart failure/LV dysfunction showing the worse prognosis and myocardial infarction with normal coronary arteries, the best prognosis.[4] Follow-up evaluations are targeted at identifying patients at increased risk for adverse cardiac outcome (heart failure, arrhythmias, or cardiac death), or identifying underlying disease processes which might guide further management (eg, genetic counseling and cascade screening in patients with underlying P/LP variants, immunosuppression in specific cardiomyopathies or associated autoimmune conditions). Figure 6 gives an overview of potential guidance on follow-up investigations.

Follow-Up Imaging

Imaging at follow-up can serve a multifold purpose. (1) Imaging can be used to identify patients with LV-dysfunction, who need guideline directed medical therapy. Data from the Lombardy registry indicate that at follow-up (196 days [interquartile range=126–349 days]) only 1% of patients with uncomplicated (thus LVEF >50% in acute phase) developed a LVEF <50%. In contrary a much higher percentage (14.5%) of patients with fulminant or complicated myocarditis had a persistently reduced LVEF <50%.[19] (2) Imaging can be used to further assess the prognosis. Data from the ITAMY (Italian Study in Myocarditis) registry showed in 182 patients with a repeat CMR, in 10% of patients a complete resolution and in 46% a reduction in LGE, while 14% had an increase in LGE. An increase in LGE or LGE at follow-up without edema were associated with a higher risk of major cardiac events.[25] This indicates that LGE in the acute setting is not fixed and can resolve. Furthermore, the absence of worse prognosis in patients with LGE and ongoing edema might suggest active inflammation with a further capability to recover. (3) Imaging such as FDG-PET can be useful for the detection of cardiac sarcoidosis and the response to therapy in cardiac sarcoidosis. The ongoing STREAM study (FDG-PET/CT Images Comparing to MRI and Endomyocardial Biopsy in Myocarditis) is evaluating if FDG-PET offers diagnostic improvement when added to CMR, a feature which might be useful in patients with probable myocarditis (Figure 3).[34]

Follow-Up Arrhythmia Monitoring

Myocarditis is a pro-arrhythmogenic condition. In acute myocarditis, ventricular arrhythmias are often polymorphic and relate to inflammation induced ion channel dysfunction and altered intracellular signaling. In the setting of a chronic inflammatory cardiomyopathy, VAs are often monomorphic and relate to scar re-entry circuits.[35] Several factors determine the risk for VAs in patients with myocarditis, including VAs during the initial admission, the underlying inflammatory infiltrate (highest risk in giant cell myocarditis), extent and location of LGE (more in the case of mid-wall LGE) and the LVEF on the initial CMR as well as the extent of areas with low potentials during bipolar endocardial voltage mapping.[13,26,35] As acute inflammation often completely resolves, the risk for VAs might also dissipate. However, because of the development of scar, scar-related monomorphic VT can develop which are often suitable for VT ablation. In patients receiving immunosuppressive therapy, the risk of ventricular arrhythmia also diminishes.[36] But no adequte powered randomized controlled trials have evaluated the use of immunosuppressive therapy for ventricular arrhythmia management. Holter monitoring and 12-lead ECG assessment within the first months after discharge combined with CMR assessment at 3 months helps to identify patients at high risk of ventricular arrhythmia and in need for implantable cardioverter defibrillator implantation.

Follow-Up Exercise Testing

There is general consensus that patients diagnosed with myocarditis should be restricted from exercise

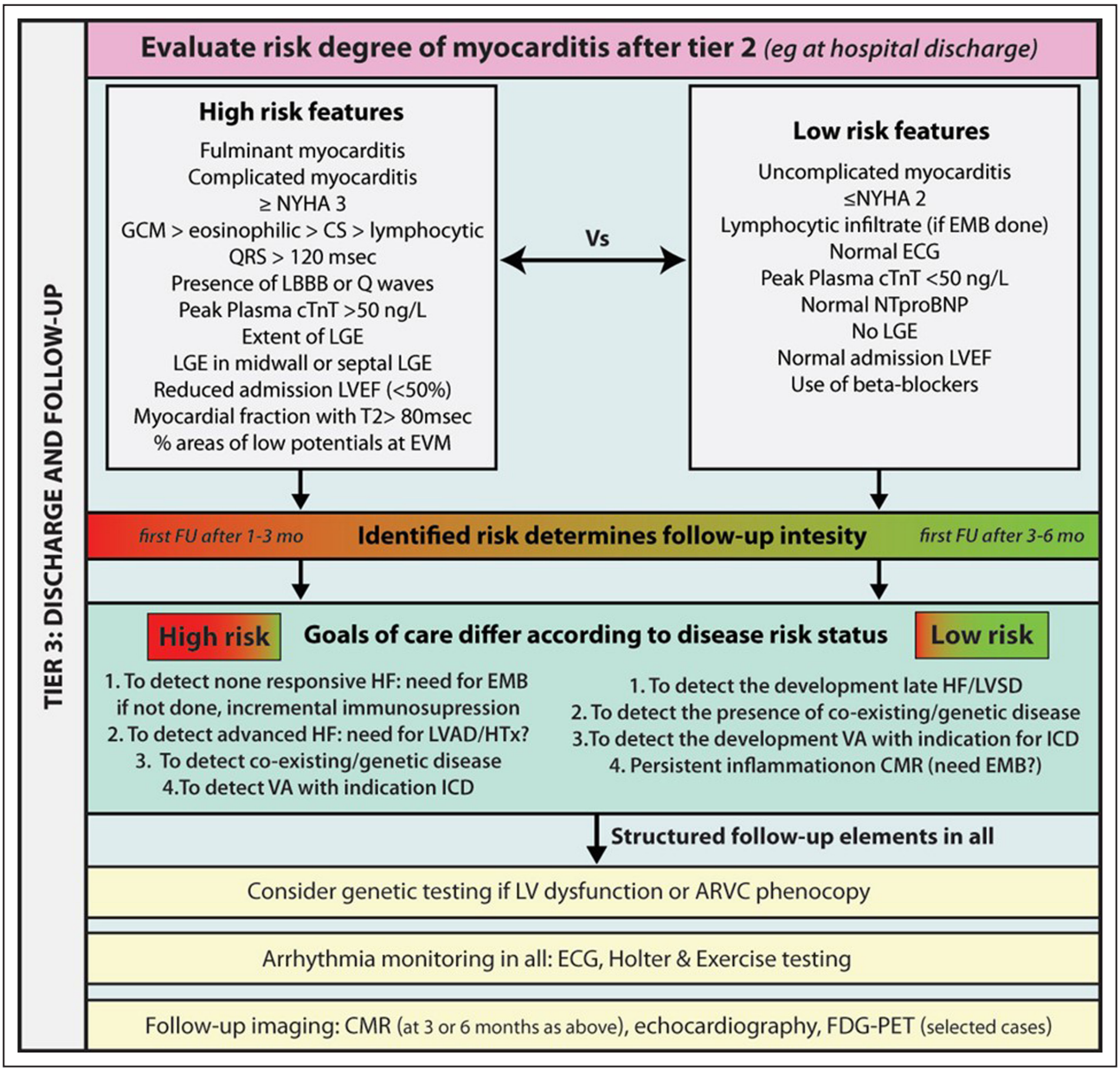

Figure 6. Tier 3—post-diagnosis investigations and care.
ARVC indicates arrhythmogenic right ventricular cardiomyopathy; CMR, cardiac magnetic resonance; CS, cardiac sarcoidosis; EVM, endocardial voltage mapping; FDG-PET, fluroredeoxyglucose positron emission tomography; FU, follow-up; GCM, giant cell myocarditis; GDMT, guideline-directed medical therapy; HF, heart failure; HTX, heart transplant; ICD, implantable cardioverter-defibrillator; LBBB, left bundle branch block; LGE, late gadolinium enhancement; LVAD, left ventricular assist device; LVSD, left ventricular systolic dysfunction; and NYHA, New York Heart Association Class.

programs and competitive sports for a period of 3 to 6 months (longer abstinence time may be needed in the presence of high-risk features). Return to play after myocarditis is a particular important subject in young athletes, as myocarditis is a common reason for sudden death in young athletes.[37] Additionally myocarditis and arrhythmogenic cardiomyopathy can share a similar genetic background which manifest with worse phenotype in the presence of endurance sport.[38,39] While physician should refrain from performing exercise testing in the acute phase of myocarditis, a recent position paper from the European Association of Preventive Cardiology, suggests to perform exercise testing at 3 to 6 months follow-up.[37] Clinically relevant arrhythmias, such as frequent or complex repetitive forms of ventricular or supraventricular arrhythmias on exercise testing at follow-up should refrain the re-initiation of competitive sports. Athletes should undergo periodical reassessment, particularly within the first 2 years and yearly thereafter (if LGE present) and

can continue sports if LVEF remains normal, myocardial injury biomarkers have normalized, and no complex arrhythmias are found on Holter monitoring and exercise testing.

Genetic Evaluation

As illustrated in Figure 1, the genetic background can predispose to the development of myocarditis, both through human leukocyte antigen haplotypes[40] that might predispose to immune polarization (eg, concomitant auto-immune disorder) or by the presence of P/LP variants that generate a second hit to the myocardium. Two cohorts of adult patients with EMB proven myocarditis show that between 16% and 31% of patients had P/LP variants, a percentage significantly higher in comparison with a matched cohort without myocarditis.[7,41] In both cohorts TTN (43%–73%) and desmoplakin (DSP) (9%–13%) variants were the most common variants in addition to other arrhythmogenic cardiomyopathy genes. The overlap between the arrhythmogenic cardiomyopathy gene DSP and myocarditis is well recognized, leading to episode of exercise induced myocarditis episodes.[38,39] Inhibition of the canonical Wnt–β-catenin signaling (occurring in DSP mutation), is associated with Th2 immune polarization, potentially explaining predisposition to myocarditis.[42] The prevalence of P/LP variants in myocarditis is in the range of variants found in isolated dilated cardiomyopathy (around 10%–25%) and little lower than in familial dilated cardiomyopathy (25%–40%).[43] The American College of Medical Genetics and Genomics recommend genetic testing at the time of new cardiomyopathy diagnosis.[43] As such genetic screening seems appropriate in myocarditis cases with LV dysfunction or patients with normal LV function but a CMR compatible with arrhythmogenic cardiomyopathy. Findings of P/LP variants carry important consequences such as genetic counseling (including preimplantation counseling as many patients are of conceptive age), cascade screening, persistent prohibition of competitive sports or prohibition of down-titration of guideline directed medical therapy in patients with recovered ejection fraction.[44] Currently no adult cases of myocarditis with severe P/LP variants such as lamin A/C are reported, that would also influence decisions towards implantable cardioverter defibrillator implantation.

Management

Although a detailed description on the management of myocarditis is beyond the scope of this article. The tier-based evaluation allows to identify mechanisms of risk (heart failure, arrhythmia, persistent inflammation, P/LP variants) that could warrant altered management as reflected in Figure S1.

CONCLUSIONS

Myocarditis is a heterogeneous disease in terms of disease severity and manifestation. As many patients with suspected myocarditis will present in different subspecialties of cardiology, well organized myocarditis care pathways spanning beyond the boundaries of subspecialty cardiology disciplines are advisable. Using a 3-tier diagnostic approach allows for the structured evaluation of the patients with suspected myocarditis allowing to titrate the diagnostic and therapeutic process to the patients' individualized needs. Such a structured tier approach can easily be supplemented by emerging advances in the future, such as immune phenotyping that could potentially shape the diagnosis, classification, and therapeutic basis of myocarditis.

ARTICLE INFORMATION

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Supplemental Material

Data S1
Tables S1–S2
Figure S1
References [45–66]

REFERENCES

1. Bang V, Ganatra S, Shah SP, Dani SS, Neilan TG, Thavendiranathan P, Resnic FS, Piemonte TC, Barac A, Patel R, et al. Management of patients with giant cell myocarditis: JACC review topic of the week. *J Am Coll Cardiol*. 2021;77:1122–1134. doi: 10.1016/j.jacc.2020.11.074
2. Trivieri MG, Spagnolo P, Birnie D, Liu P, Drake W, Kovacic JC, Baughman R, Fayad ZA, Judson MA. Challenges in cardiac and pulmonary sarcoidosis: JACC state-of-the-art review. *J Am Coll Cardiol*. 2020;76:1878–1901. doi: 10.1016/j.jacc.2020.08.042
3. Cooper LT Jr. Myocarditis. *N Engl J Med*. 2009;360:1526–1538. doi: 10.1056/NEJMra0800028
4. Caforio AL, Calabrese F, Angelini A, Tona F, Vinci A, Bottaro S, Ramondo A, Carturan E, Iliceto S, Thiene G, et al. A prospective study of biopsy-proven myocarditis: prognostic relevance of clinical and aetiopathogenetic features at diagnosis. *Eur Heart J*. 2007;28:1326–1333. doi: 10.1093/eurheartj/ehm076
5. Gil-Cruz C, Perez-Shibayama C, De MA, Ronchi F, van der Borght K, Niederer R, Onder L, Lutge M, Novkovic M, Nindl V, et al. Microbiota-derived peptide mimics drive lethal inflammatory cardiomyopathy. *Science*. 2019;366:881–886. doi: 10.1126/science.aav3487

6. Myers JM, Cooper LT, Kem DC, Stavrakis S, Kosanke SD, Shevach EM, Fairweather D, Stoner JA, Cox CJ, Cunningham MW. Cardiac myosin-Th17 responses promote heart failure in human myocarditis. *JCI Insight*. 2016;1:1. doi: 10.1172/jci.insight.85851
7. Artico J, Merlo M, Delcaro G, Cannata A, Gentile P, De AG, Paldino A, Bussani R, Ferro MD, Sinagra G. Lymphocytic myocarditis: a genetically predisposed disease? *J Am Coll Cardiol*. 2020;75:3098–3100. doi: 10.1016/j.jacc.2020.04.048
8. Poller W, Haas J, Klingel K, Kuhnisch J, Gast M, Kaya Z, Escher F, Kayvanpour E, Degener F, Opgen-Rhein B, et al. Familial recurrent myocarditis triggered by exercise in patients with a truncating variant of the desmoplakin gene. *J Am Heart Assoc*. 2020;9:e015289. doi: 10.1161/JAHA.119.015289
9. Patriki D, Gresser E, Manka R, Emmert MY, Luscher TF, Heidecker B. Approximation of the incidence of myocarditis by systematic screening with cardiac magnetic resonance imaging. *JACC Heart Fail*. 2018;6:573–579. doi: 10.1016/j.jchf.2018.03.002
10. Pasupathy S, Air T, Dreyer RP, Tavella R, Beltrame JF. Systematic review of patients presenting with suspected myocardial infarction and nonobstructive coronary arteries. *Circulation*. 2015;131:861–870. doi: 10.1161/CIRCULATIONAHA.114.011201
11. Andreini D, Dello RA, Pontone G, Mushtaq S, Conte E, Perchinunno M, Guglielmo M, Coutinho SA, Magatelli M, Baggiano A, et al. CMR for identifying the substrate of ventricular arrhythmia in patients with normal echocardiography. *JACC Cardiovasc Imaging*. 2020;13:410–421. doi: 10.1016/j.jcmg.2019.04.023
12. Lakkireddy D, Turagam MK, Yarlagadda B, Dar T, Hamblin M, Krause M, Parikh V, Bommana S, Atkins D, Di BL, et al. Myocarditis causing premature ventricular contractions: insights from the MAVERIC registry. *Circ Arrhythm Electrophysiol*. 2019;12:e007520. doi: 10.1161/CIRCEP.119.007520
13. Peretto G, Sala S, Basso C, Rizzo S, Radinovic A, Frontera A, Limite LR, Paglino G, Bisceglia C, De LG, et al. Inflammation as a predictor of recurrent ventricular tachycardia after ablation in patients with myocarditis. *J Am Coll Cardiol*. 2020;76:1644–1656. doi: 10.1016/j.jacc.2020.08.012
14. Caforio AL, Pankuweit S, Arbustini E, Basso C, Gimeno-Blanes J, Felix SB, Fu M, Helio T, Heymans S, Jahns R, et al. Current state of knowledge on aetiology, diagnosis, management, and therapy of myocarditis: a position statement of the European Society of Cardiology Working Group on Myocardial and Pericardial Diseases. *Eur Heart J*. 2013;34:2636–2648. doi: 10.1093/eurheartj/eht210
15. Lieberman EB, Hutchins GM, Herskowitz A, Rose NR, Baughman KL. Clinicopathologic description of myocarditis. *J Am Coll Cardiol*. 1991;18:1617–1626. doi: 10.1016/0735-1097(91)90493-S
16. Olsen EG. Histopathologic aspects of viral myocarditis and its diagnostic criteria. *Heart Vessels Suppl*. 1985;1:130–132. doi: 10.1007/BF02072379
17. Ammirati E, Veronese G, Brambatti M, Merlo M, Cipriani M, Potena L, Sormani P, Aoki T, Sugimura K, Sawamura A, et al. Fulminant versus acute nonfulminant myocarditis in patients with left ventricular systolic dysfunction. *J Am Coll Cardiol*. 2019;74:299–311. doi: 10.1016/j.jacc.2019.04.063
18. McCarthy RE III, Boehmer JP, Hruban RH, Hutchins GM, Kasper EK, Hare JM, Baughman KL. Long-term outcome of fulminant myocarditis as compared with acute (nonfulminant) myocarditis. *N Engl J Med*. 2000;342:690–695. doi: 10.1056/NEJM200003093421003
19. Ammirati E, Cipriani M, Moro C, Raineri C, Pini D, Sormani P, Mantovani R, Varrenti M, Pedrotti P, Conca C, et al. Clinical presentation and outcome in a contemporary cohort of patients with acute myocarditis: multicenter Lombardy registry. *Circulation*. 2018;138:1088–1099. doi: 10.1161/CIRCULATIONAHA.118.035319
20. Johnson DB, Balko JM, Compton ML, Chalkias S, Gorham J, Xu Y, Hicks M, Puzanov I, Alexander MR, Bloomer TL, et al. Fulminant myocarditis with combination immune checkpoint blockade. *N Engl J Med*. 2016;375:1749–1755. doi: 10.1056/NEJMoa1609214
21. Blanco-Dominguez R, Sanchez-Diaz R, de la Fuente H, Jimenez-Borreguero LJ, Matesanz-Marin A, Relano M, Jimenez-Alejandre R, Linillos-Pradillo B, Tsilingiri K, et al. A novel circulating microRNA for the detection of acute myocarditis. *N Engl J Med*. 2021;384:2014–2027. doi: 10.1056/NEJMoa2003608
22. Greulich S, Kindermann I, Schumm J, Perne A, Birkmeier S, Grun S, Ong P, Schaufele T, Klingel K, Schneider S, et al. Predictors of outcome in patients with parvovirus B19 positive endomyocardial biopsy. *Clin Res Cardiol*. 2016;105:37–52. doi: 10.1007/s00392-015-0884-6
23. Kindermann I, Kindermann M, Kandolf R, Klingel K, Bultmann B, Muller T, Lindinger A, Bohm M. Predictors of outcome in patients with suspected myocarditis. *Circulation*. 2008;118:639–648. doi: 10.1161/CIRCULATIONAHA.108.769489
24. Bozkurt B, Kamat I, Hotez PJ. Myocarditis with COVID-19 mRNA vaccines. *Circulation*. 2021;144:471–484. doi: 10.1161/CIRCULATIONAHA.121.056135
25. Aquaro GD, Perfetti M, Camastra G, Monti L, Dellegrottaglie S, Moro C, Pepe A, Todiere G, Lanzillo C, Scatteia A, et al. Cardiac MR with late gadolinium enhancement in acute myocarditis with preserved systolic function: ITAMY study. *J Am Coll Cardiol*. 2017;70:1977–1987. doi: 10.1016/j.jacc.2017.08.044
26. Nordenswan HK, Lehtonen J, Ekstrom K, Raisanen-Sokolowski A, Mayranpaa MI, Vihinen T, Miettinen H, Kaikkonen K, Haataja P, Kerola T, et al. Manifestations and outcome of cardiac sarcoidosis and idiopathic giant cell myocarditis by 25-year nationwide cohorts. *J Am Heart Assoc*. 2021;10:e019415. doi: 10.1161/JAHA.120.019415
27. Seferovic PM, Tsutsui H, McNamara DM, Ristic AD, Basso C, Bozkurt B, Cooper LT Jr, Filippatos G, Ide T, Inomata T, et al. Heart failure association of the ESC, Heart Failure Society of America and Japanese Heart Failure Society position statement on endomyocardial biopsy. *Eur J Heart Fail*. 2021;23:854–871. doi: 10.1002/ejhf.2190
28. Katzmann JL, Schlattmann P, Rigopoulos AG, Noutsias E, Bigalke B, Pauschinger M, Tschope C, Sedding D, Schulze PC, Noutsias M. Meta-analysis on the immunohistological detection of inflammatory cardiomyopathy in endomyocardial biopsies. *Heart Fail Rev*. 2020;25:277–294. doi: 10.1007/s10741-019-09835-9
29. Singh V, Mendirichaga R, Savani GT, Rodriguez A, Blumer V, Elmariah S, Inglessis-Azuaje I, Palacios I. Comparison of utilization trends, indications, and complications of endomyocardial biopsy in native versus donor hearts (from the Nationwide Inpatient Sample 2002 to 2014). *Am J Cardiol*. 2018;121:356–363. doi: 10.1016/j.amjcard.2017.10.021
30. Ferreira VM, Schulz-Menger J, Holmvang G, Kramer CM, Carbone I, Sechtem U, Kindermann I, Gutberlet M, Cooper LT, Liu P, et al. Cardiovascular magnetic resonance in nonischemic myocardial inflammation: expert recommendations. *J Am Coll Cardiol*. 2018;72:3158–3176. doi: 10.1016/j.jacc.2018.09.072
31. Francone M, Chimenti C, Galea N, Scopelliti F, Verardo R, Galea R, Carbone I, Catalano C, Fedele F, Frustaci A. CMR sensitivity varies with clinical presentation and extent of cell necrosis in biopsy-proven acute myocarditis. *JACC Cardiovasc Imaging*. 2014;7:254–263. doi: 10.1016/j.jcmg.2013.10.011
32. Nadjiri J, Nieberler H, Hendrich E, Greiser A, Will A, Martinoff S, Hadamitzky M. Performance of native and contrast-enhanced T1 mapping to detect myocardial damage in patients with suspected myocarditis: a head-to-head comparison of different cardiovascular magnetic resonance techniques. *Int J Cardiovasc Imaging*. 2017;33:539–547. doi: 10.1007/s10554-016-1029-3
33. Spieker M, Haberkorn S, Gastl M, Behm P, Katsianos S, Horn P, Jacoby C, Schnackenburg B, Reinecke P, Kelm M, et al. Abnormal T2 mapping cardiovascular magnetic resonance correlates with adverse clinical outcome in patients with suspected acute myocarditis. *J Cardiovasc Magn Reson*. 2017;19:38. doi: 10.1186/s12968-017-0350-x
34. Ozieranski K, Tyminska A, Kobylecka M, Caforio ALP, Sobic-Saranovic D, Ristic AD, Maksimovic R, Seferovic PM, Marcolongo R, Krolicki L, et al. Positron emission tomography in clinically suspected myocarditis—STREAM study design. *Int J Cardiol*. 2021;332:113–118. doi: 10.1016/j.ijcard.2021.02.068
35. Peretto G, Sala S, Rizzo S, Palmisano A, Esposito A, De CF, Campochiaro C, De LG, Foppoli L, Dagna L, et al. Ventricular arrhythmias in myocarditis: characterization and relationships with myocardial inflammation. *J Am Coll Cardiol*. 2020;75:1046–1057. doi: 10.1016/j.jacc.2020.01.036
36. Peretto G, Sala S, De LG, Marcolongo R, Campochiaro C, Sartorelli S, Tresoldi M, Foppoli L, Palmisano A, Esposito A, et al. Immunosuppressive therapy and risk stratification of patients with myocarditis presenting with ventricular arrhythmias. *JACC Clin Electrophysiol*. 2020;6:1221–1234. doi: 10.1016/j.jacep.2020.05.013
37. Pelliccia A, Solberg EE, Papadakis M, Adami PE, Biffi A, Caselli S, La GA, Niebauer J, Pressler A, Schmied CM, et al. Recommendations for participation in competitive and leisure time sport in athletes with cardiomyopathies, myocarditis, and pericarditis: position statement of the Sport Cardiology Section of the European Association of Preventive Cardiology (EAPC). *Eur Heart J*. 2019;40:19–33. doi: 10.1093/eurheartj/ehy730

38. Alley R, Grizzard JD, Rao K, Markley R, Trankle CR. Inflammatory episodes of desmoplakin cardiomyopathy masquerading as myocarditis: unique features on cardiac magnetic resonance imaging. *JACC Cardiovasc Imaging*. 2021;14:1466–1469. doi: 10.1016/j.jcmg.2020.07.028
39. Reichl K, Kreykes SE, Martin CM, Shenoy C. Desmoplakin variant-associated arrhythmogenic cardiomyopathy presenting as acute myocarditis. *Circ Genom Precis Med*. 2018;11:e002373. doi: 10.1161/CIRCGEN.118.002373
40. Lozano MD, Rubocki RJ, Wilson JE, McManus BM, Wisecarver JL. Human leukocyte antigen class II associations in patients with idiopathic dilated cardiomyopathy. Myocarditis Treatment Trial Investigators. *J Card Fail*. 1997;3:97–103. doi: 10.1016/S1071-9164(97)90041-5
41. Kontorovich AR, Patel N, Moscati A, Richter F, Peter I, Purevjav E, Selejan SR, Kindermann I, Towbin JA, Bohm M, et al. Myopathic cardiac genotypes increase risk for myocarditis. *JACC Basic Transl Sci*. 2021;6:584–592. doi: 10.1016/j.jacbts.2021.06.001
42. Li X, Xiang Y, Li F, Yin C, Li B, Ke X. WNT/beta-catenin signaling pathway regulating T cell-inflammation in the tumor microenvironment. *Front Immunol*. 2019;10:2293. doi: 10.3389/fimmu.2019.02293
43. Hershberger RE, Givertz MM, Ho CY, Judge DP, Kantor PF, McBride KL, Morales A, Taylor MRG, Vatta M, Ware SM. Genetic evaluation of cardiomyopathy: a clinical practice resource of the American College of Medical Genetics and Genomics (ACMG). *Genet Med*. 2018;20:899–909. doi: 10.1038/s41436-018-0039-z
44. Halliday BP, Wassall R, Lota AS, Khalique Z, Gregson J, Newsome S, Jackson R, Rahneva T, Wage R, Smith G, et al. Withdrawal of pharmacological treatment for heart failure in patients with recovered dilated cardiomyopathy (TRED-HF): an open-label, pilot, randomised trial. *Lancet*. 2019;393:61–73. doi: 10.1016/S0140-6736(18)32484-X
45. Perniola R. Expression of the autoimmune regulator gene and its relevance to the mechanisms of central and peripheral tolerance. *Clin Dev Immunol*. 2012;2012:207403. doi: 10.1155/2012/207403
46. Lv H, Lipes MA. Role of impaired central tolerance to alpha-myosin in inflammatory heart disease. *Trends Cardiovasc Med*. 2012;22:113–117. doi: 10.1016/j.tcm.2012.07.005
47. Sakaguchi S, Yamaguchi T, Nomura T, Ono M. Regulatory T cells and immune tolerance. *Cell*. 2008;133:775–787. doi: 10.1016/j.cell.2008.05.009
48. Li Y, Heuser JS, Kosanke SD, Hemric M, Cunningham MW. Cryptic epitope identified in rat and human cardiac myosin S2 region induces myocarditis in the Lewis rat. *J Immunol*. 2004;172:3225–3234. doi: 10.4049/jimmunol.172.5.3225
49. Zhang P, Cox CJ, Alvarez KM, Cunningham MW. Cutting edge: cardiac myosin activates innate immune responses through TLRs. *J Immunol*. 2009;183:27–31. doi: 10.4049/jimmunol.0800861
50. Donermeyer DL, Beisel KW, Allen PM, Smith SC. Myocarditis-inducing epitope of myosin binds constitutively and stably to I-Ak on antigen-presenting cells in the heart. *J Exp Med*. 1995;182:1291–1300. doi: 10.1084/jem.182.5.1291
51. Liu Y, Zhu H, Su Z, Sun C, Yin J, Yuan H, Sandoghchian S, Jiao Z, Wang S, Xu H. IL-17 contributes to cardiac fibrosis following experimental autoimmune myocarditis by a PKCbeta/Erk1/2/NF-kappaB-dependent signaling pathway. *Int Immunol*. 2012;24:605–612. doi: 10.1093/intimm/dxs056
52. Wei L. Immunological aspect of cardiac remodeling: T lymphocyte subsets in inflammation-mediated cardiac fibrosis. *Exp Mol Pathol*. 2011;90:74–78. doi: 10.1016/j.yexmp.2010.10.004
53. Chen P, Baldeviano GC, Ligons DL, Talor MV, Barin JG, Rose NR, Cihakova D. Susceptibility to autoimmune myocarditis is associated with intrinsic differences in CD4(+) T cells. *Clin Exp Immunol*. 2012;169:79–88. doi: 10.1111/j.1365-2249.2012.04598.x
54. Martinez NE, Sato F, Kawai E, Omura S, Chervenak RP, Tsunoda I. Regulatory T cells and Th17 cells in viral infections: implications for multiple sclerosis and myocarditis. *Future Virol*. 2012;7:593–608. doi: 10.2217/fvl.12.44
55. An B, Liu X, Li G, Yuan H. Interleukin-37 ameliorates coxsackievirus B3-induced viral myocarditis by modulating the Th17/regulatory T cell immune response. *J Cardiovasc Pharmacol*. 2017;69:305–313. doi: 10.1097/FJC.0000000000000476
56. Martin P, Blanco-Dominguez R, Sanchez-Diaz R. Novel human immunomodulatory T cell receptors and their double-edged potential in autoimmunity, cardiovascular disease and cancer. *Cell Mol Immunol*. 2021;18:919–935. doi: 10.1038/s41423-020-00586-4
57. Cruz-Adalia A, Jimenez-Borreguero LJ, Ramirez-Huesca M, Chico-Calero I, Barreiro O, Lopez-Conesa E, Fresno M, Sanchez-Madrid F, Martin P. CD69 limits the severity of cardiomyopathy after autoimmune myocarditis. *Circulation*. 2010;122:1396–1404. doi: 10.1161/CIRCULATIONAHA.110.952820
58. Chen X, Zeng XH, Wang M, Chen L, Zhang N, Rao M, Yang PC, Song J. Bcl2-like protein 12 is required for the aberrant T helper-2 polarization in the heart by enhancing interleukin-4 expression and compromising apoptotic machinery in $CD4^+$ T cells. *Circulation*. 2018;138:2559–2568. doi: 10.1161/CIRCULATIONAHA.118.033890
59. Maier E, Duschl A, Horejs-Hoeck J. STAT6-dependent and -independent mechanisms in Th2 polarization. *Eur J Immunol*. 2012;42:2827–2833. doi: 10.1002/eji.201242433
60. Corsten MF, Papageorgiou A, Verhesen W, Carai P, Lindow M, Obad S, Summer G, Coort SL, Hazebroek M, van Leeuwen R, et al. MicroRNA profiling identifies microRNA-155 as an adverse mediator of cardiac injury and dysfunction during acute viral myocarditis. *Circ Res*. 2012;111:415–425. doi: 10.1161/CIRCRESAHA.112.267443
61. Heymans S, Corsten MF, Verhesen W, Carai P, van Leeuwen RE, Custers K, Peters T, Hazebroek M, Stoger L, Wijnands E, et al. Macrophage microRNA-155 promotes cardiac hypertrophy and failure. *Circulation*. 2013;128:1420–1432. doi: 10.1161/CIRCULATIONAHA.112.001357
62. Xu HF, Ding YJ, Zhang ZX, Wang ZF, Luo CL, Li BX, Shen YW, Tao LY, Zhao ZQ. MicroRNA21 regulation of the progression of viral myocarditis to dilated cardiomyopathy. *Mol Med Rep*. 2014;10:161–168. doi: 10.3892/mmr.2014.2205
63. Chang JC, Wooten EC, Tsimelzon A, Hilsenbeck SG, Gutierrez MC, Tham YL, Kalidas M, Elledge R, Mohsin S, Osborne CK, et al. Patterns of resistance and incomplete response to docetaxel by gene expression profiling in breast cancer patients. *J Clin Oncol*. 2005;23:1169–1177. doi: 10.1200/JCO.2005.03.156
64. Gruosso T, Gigoux M, Manem VSK, Bertos N, Zuo D, Perlitch I, Saleh SMI, Zhao H, Souleimanova M, Johnson RM, et al. Spatially distinct tumor immune microenvironments stratify triple-negative breast cancers. *J Clin Invest*. 2019;129:1785–1800. doi: 10.1172/JCI96313
65. Halloran PF, Potena L, Van Huyen JD, Bruneval P, Leone O, Kim DH, Jouven X, Reeve J, Loupy A. Building a tissue-based molecular diagnostic system in heart transplant rejection: the heart molecular microscope diagnostic (MMDx) system. *J Heart Lung Transplant*. 2017;36:1192–1200. doi: 10.1016/j.healun.2017.05.029
66. Orr C, Najm A, Biniecka M, McGarry T, Ng CT, Young F, Fearon U, Veale DJ. Synovial immunophenotype and anti-citrullinated peptide antibodies in rheumatoid arthritis patients: relationship to treatment response and radiologic prognosis. *Arthritis Rheumatol*. 2017;69:2114–2123. doi: 10.1002/art.40218