Prevalence of Pathogenic Variants in Cardiomyopathy-Associated Genes in Acute Myocarditis: A Systematic Review and Meta-Analysis

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Running Title: Genetic Architecture of Acute Myocarditis

Funding: The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Disclosures: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Abstract

Background: Acute myocarditis is an inflammatory condition that may precede the development of dilated or arrhythmogenic cardiomyopathy.

Objectives: To investigate the reported prevalence of pathogenic/likely pathogenic (P/LP) variants in cardiomyopathy-associated genes in patients with acute myocarditis.

Methods: For this systematic review and meta-analysis, we searched Pubmed and Embase databases on March 04, 2023. Observational studies evaluating the prevalence of P/LP variants in cardiomyopathy-associated genes in patients with acute myocarditis were included. Studies were stratified into adult and pediatric age groups and for the following scenarios: (a) complicated myocarditis (i.e., presenting with acute heart failure, reduced left ventricular ejection fraction, or life-threatening ventricular arrhythmias); (b) uncomplicated myocarditis. The study was registered on PROSPERO (CRD42023408668) and followed PRISMA guidelines.

Results: Of 732 studies identified, 8 met the inclusion criteria, providing data for 586 patients with acute myocarditis. A total of 89 P/LP variants in cardiomyopathy-associated genes were reported in 85 patients. In uncomplicated myocarditis the pooled prevalence was 4.2% (95% CI 1.8-7.4%, I^2 1.4%), while in complicated myocarditis pooled prevalence was 21.9% (95% CI 14.3-30.5%, I^2 38.8%) and 44.5% (95% CI 22.7-67.4%, I^2 52.8%) in adults and children, respectively. P/LP variants in desmosomal genes were predominant in uncomplicated myocarditis (64%), while sarcomeric gene variants were more prevalent in complicated myocarditis (58% in adults and 71% in children).

Conclusions: Genetic variants are present in a large proportion of patients with acute myocarditis. The prevalence of genetic variants and the genes involved vary according to age and clinical presentation.

Keywords: myocarditis; genetics; cardiomyopathy.

Abbreviations

ACM = arrhythmogenic cardiomyopathy

DCM = dilated cardiomyopathy

LVEF = left ventricular ejection fraction

P/LP = pathogenic/likely pathogenic

SCD = sudden cardiac death

Introduction

Myocarditis is an inflammatory disease of the myocardium which results from a wide range of infectious and non-infectious causes[1]. Myocarditis has different clinical presentations according to the extent of cardiac involvement[2] and, while spontaneous recovery occurs in many patients, some progress to chronic ventricular dysfunction[3].

The observation that acute myocarditis can be the first clinical manifestation in patients who develop arrhythmogenic cardiomyopathy (ACM) has led to the hypothesis that pathological variants in structural genes might increase myocardial vulnerability to inflammation induced by infective agents or other stressors[4]. Following the description of myocarditis-related sudden cardiac death (SCD) in individuals who were found to be carriers of disease-causing mutations in desmosomal genes[5], other case reports, case series, and cohort studies have reported the presence of pathological variants in cardiomyopathy-associated genes among patients with acute myocarditis[6–10]. Other data suggest that genetic variation may play a role in determining the clinical course and outcomes of patients with acute myocarditis[2].

The aim of this systematic review and meta-analysis was to define the prevalence of pathogenic/likely pathogenic (P/LP) variants in cardiomyopathy-associated genes in patients with acute myocarditis.

Methods

This systematic review and meta-analysis is reported in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) statement[11]. The review was registered on PROSPERO (CRD42023408668). The MOOSE (Meta-analyses of Observational Studies in Epidemiology) checklist[12] is reported as **Supplemental Table 1**.

Eligibility Criteria

Criteria for inclusion in the study were: 1) observational full-length original article or

research letter describing cohorts involving pediatric and/or adult individuals with a diagnosis of acute myocarditis; 2) reported data on screening for cardiomyopathy-associated genes; 3) English language studies. Studies published as abstract, case report, review, preprint article or preclinical studies were excluded. Studies including patients with acute myocarditis in the context of additional criteria suggestive for inherited cardiomyopathies (e.g., family history of cardiomyopathy or SCD, or right ventricular involvement) were excluded.

The study cohorts were stratified into adult (≥18 years) and pediatric age (<18 years) groups and for the following clinical scenarios: (1) complicated myocarditis (i.e., presentation with acute heart failure, reduced left ventricular ejection fraction [LVEF], or life-threatening ventricular arrhythmias); (2) uncomplicated myocarditis, according to the definition of each individual study (**Table 1**).

Information sources and search strategy

We performed a systematic review of published studies searching the Pubmed and Embase databases on March 04, 2023, using keywords pertaining to myocarditis and genetic testing. The search strategy is presented in **Supplemental Table 2**. As per eligibility criteria, the strategy was limited to observational full-length original articles or research letters and English language studies. In addition, reference lists of the articles included in the review were manually screened to identify additional studies.

Selection and data collection process

Two authors (E.M. and A.B.) independently screened the studies, obtained the complete reports of potentially relevant studies, and reviewed each paper using predefined eligibility criteria. The process of study selection is reported in **Figure 1**. The same authors (E.M. and A.B.) independently collected the data from each study. Controversies about study selection or data collection were resolved through discussion between the two authors (E.M. and A.B.).

Outcomes and data items

The outcome was the prevalence of P/LP variants in cardiomyopathy-associated genes among patients with acute myocarditis. For each reported study, the following information was collected: first author; date of publication and country; study design; number of individuals undergoing genetic testing; methods of genetic testing; number of patients showing P/LP variants in cardiomyopathy-associated genes; P/LP variants in cardiomyopathy-associated genes identified; and clinical data.

Genetic variants were classified as P, LP, variant of unknown significance, likely benign or benign using the American College of Medical Genetics and Genomics (ACMG) classification[13].

Study risk of bias assessment

Two independent authors (E.M. and A.B.) evaluated the risk of bias using the Joanna Briggs Institute (JBI) critical appraisal checklist for studies reporting prevalence data[14]. Any discrepancies in judgements of risk of bias were solved through discussion between the two authors (E.M. and A.B.).

Synthesis methods

The prevalence of P/LP variants in cardiomyopathy-associated genes was calculated as the ratio between patients with P/LP variants and the screened populations. Specifically, we synthesized eligible studies and derived the pooled estimate for the prevalence of P/LP variants in cardiomyopathy-associated genes within each clinical scenario. The 95% confidence intervals (CI) of individual studies were calculated using the exact binomial (Clopper-Pearson) method. The mean prevalence and CI of individual studies were reported. Heterogeneity was quantified using the I-squared measure. Publication bias was assessed for using Egger's regression-based test. Results of the Egger's test were reported as t-value and p-value (2-tailed). Synthesis methods were performed with JBI SUMARI[15] and

Comprehensive Meta-analysis V4.

Results

We identified 732 articles through the initial database search. We removed 443 non-human (n = 97) or non-English records (n = 20), or non-research articles/letters before screening (n = 326). After removing 276 records based on the title and abstract, we retrieved 13 articles for full-text review. Of these, three were excluded as genetic testing was not systematically performed in patients with myocarditis[16–18], and two were excluded due to the presence of additional diagnostic criteria suggestive for inherited cardiomyopathy (i.e., presence of documented episode of acute myocarditis and at least one family member with a cardiomyopathy or a history of SCD[19]; clinically suspected myocarditis associated with sustained ventricular arrhythmias and/or right ventricular abnormalities[10]). A total of 8 studies met the inclusion criteria and were included in the systematic review and meta-analysis[6–10, 19–23].

The eight analysed studies included a total of 586 patients. There were two observational studies from the USA (125 patients)[20, 21], two from Germany (54 patients)[9, 22], and one each from Italy (36 patients)[8], the UK (336 patients)[7], Spain (28 patients)[6], and the Netherlands (7 patients)[23], respectively. **(Table 1)**. Three studies included adult populations[6–8], four studies pediatric populations[9, 20, 22, 23], and one study included both adult and paediatric patients[21]. Details of studies included in the meta-analysis are reported in **Table 1**, **Supplemental Table 3**, and **Supplemental Table 4**.

Criteria for the diagnosis of acute myocarditis varied among the different studies included. The diagnosis was biopsy-proven in three studies[8, 9, 22] and clinically suspected in one study[20], while four studies included patients with either biopsy-proven or clinically suspected diagnosis[6, 7, 21, 23]. Among studies including individuals with uncomplicated myocarditis, diagnosis of acute myocarditis was biopsy-proven or clinically suspected in all

the four studies[6, 7, 9, 21]. Among studies including children with complicated myocarditis, diagnosis was clinically suspected in one study[20], biopsy-proven in two studies[9, 22], and either biopsy-proven or clinically suspected in one study[23], while among studies including adults with complicated myocarditis, diagnosis was biopsy-proven in one study[8] and either biopsy-proven or clinically suspected in two studies[6, 7].

Pooled prevalence of P/LP variants in the overall population

The baseline characteristics are summarized in **Table 1** and **Table 2**. Among the 586 patients who underwent genetic testing, 85 carried ≥1 P/LP variant in cardiomyopathy-associated genes with a pooled prevalence in a random effects model of 24.7% (95% CI 12.3-39.4%) **(Supplemental Figure 1)**. However, due to the different study populations and inclusion criteria, significant heterogeneity was present (I^2 88.1%; t-value 2.59, p-value 0.04).

A total of 89 P/LP variants in cardiomyopathy-associated genes were identified **(Figure 2, Supplemental Table 5)**. The most common variants occurred in genes encoding for sarcomeric (n = 45, 51%) or desmosomal proteins (n = 15, 17%). Other genes affected were those encoding for sarcolemma proteins (n = 6, 7%), co-chaperone heat shock proteins (Co-Chap HSP) (n = 6, 7%), ion channels (n = 1, 1%), RNA binding proteins (n = 4, 4%), DNA binding proteins (n = 1, 1%), proteins of the cytoskeleton (n = 4, 5%), nuclear envelope (n = 5, 6%), and sarcoplasmic reticulum (n = 3, 3%). Among the sarcomeric genes, the most common variants occurred in *TTN*, followed by *TNNT2*, *MYH7*, *TNNI3*, *TNNC1*, *MYBPC3*, and *TNNT1*, while among desmosomal genes, variants occurred in *DSP*, *PKP2*, and *DSG2*.

Four studies included adult patients, providing data for 517 patients[6-8,20]. Regardless of clinical presentation, the pooled prevalence of P/LP variants in cardiomyopathy-associated genes in adults was 11.0% (95% CI 8.3-13.9%, I^2 82.1%; t-value 1.55, p-value 0.26) (**Supplemental Figure 2**).

Pooled prevalence of P/LP variants in patients with uncomplicated myocarditis

Four studies included patients with uncomplicated myocarditis, providing data for 286 patients[6, 7, 9, 21] **(Figure 3)**. The pooled prevalence of P/LP variants in cardiomyopathy-associated genes was 4.2% (95% CI 1.8-7.4%, I^2 1.4%; t-value 1.46, p-value 0.28). After excluding one study that did not report the variants identified in uncomplicated myocarditis[21], 13 P/LP variants in cardiomyopathy-associated genes were identified **(Figure 4)**. The variants occurred most frequently in desmosomal genes (n = 9, 69%), with *DSP* the most common, and in *TTN* (n = 2, 15%).

Pooled prevalence of P/LP variants in patients with complicated myocarditis

Eight studies included patients with complicated myocarditis, providing data for 209 adults (4 studies)[6–8, 21] and 47 children (4 studies)[9, 20, 22, 23] **(Figure 3)**. The pooled prevalence of P/LP variants in cardiomyopathy-associated genes was 21.9% (95% CI 14.3-30.5%, I^2 38.8%; t-value 1.88, p-value 0.20) in adults and 44.5% (95% CI 22.7-67.4%, I^2 52.8%; t-value 0.21, p-value 0.85) in children.

After excluding one study which did not report the variants identified in complicated myocarditis[21], 33 and 21 P/LP variants in cardiomyopathy-associated genes were identified in adults and children with complicated myocarditis, respectively **(Figure 4)**. Variants mostly occurred in sarcomeric genes in adults and paediatric patients (n = 19, 58% and n = 15, 71%, respectively), with *TTN* the most frequently involved gene in adults, and non-*TTN* sarcomeric genes (i.e., *MYH7*, *TNNI3*, *TNNT2*, *MYBPC3*, and *TNNC1*) representing the most prevalent in children.

Bias assessments

The risk of bias assessments for prevalence studies is summarised in **Supplemental Table 6**. The STROBE (STrengthening the Reporting of OBservational studies in Epidemiology) checklist for each study included into the systematic review and meta-analysis is reported in **Supplemental Table 7**.

Discussion

This systematic review and meta-analysis assessed the prevalence of P/LP variants in cardiomyopathy-associated genes in patients with acute myocarditis. We found that the reported frequency of P/LP variants and the genes involved vary according to age and clinical presentation.

There is a growing focus on genetic predisposition in cardiac conditions previously considered to be caused predominantly by environmental or immunological mechanisms. For example, several studies have underlined the role of rare genetic traits in pregnancy-, alcohol- and chemotherapy-induced cardiomyopathy, with the identification of pathogenic variants in cardiomyopathy-associated genes such as *TTN* or *MYH7* in a proportion of patients[24–26]. This systematic review suggests that genetic mechanisms also predispose the myocardium to acute myocarditis.

In this study, we found that a significant proportion of patients with myocarditis harbor a P/LP variant in a cardiomyopathy-associated gene. A pooled analysis of positive genetic testing across all studies, regardless of age and myocarditis severity, demonstrated a significant degree of heterogeneity, reflective of the different characteristics within the study cohorts. However, when the studies were stratified by clinical presentation this heterogeneity diminished. Specifically, patients who experienced uncomplicated myocarditis rarely carried a genetic variant (only 4% of cases) whereas the prevalence was much higher in patients with myocarditis presenting with acute heart failure, reduced LVEF, or life-threatening ventricular arrhythmias, ranging from 22% in adults to 45% in children.

Uncomplicated myocarditis

We found a significant relationship between the clinical presentation of myocarditis and the genes affected by P/LP variants. Consistently, *DSP* variants were identified mostly in patients with acute uncomplicated myocarditis but were less commonly observed in adults

and absent in children presenting with acute heart failure or ventricular arrhythmias.

DSP variants cause a distinct cardiomyopathy characterized by left ventricular fibrosis and, in advanced stages, a high incidence of ventricular arrhythmias[27]. It has been observed that in patients carrying *DSP* variants, myocarditis can be the first clinical manifestation of an underlying cardiomyopathy[28, 29] and that episodes of acute myocardial injury often occur in the presence of normal systolic function, presaging progression of the disease[27, 30].

Complicated myocarditis

Pathogenic variants in *TTN* are commonly reported in patients with DCM (up to 25% of cases) [31] and have been implicated in about 10% of patients with peripartum cardiomyopathy[24] and 12% of individuals with chemotherapy-induced cardiomyopathy[25]. In this study, we found that *TTN* variants are also common among patients with myocarditis presenting with acute heart failure, reduced LVEF, or ventricular arrhythmias. These findings are consistent with studies showing that *TTN* variants convey a predisposition to disease in the presence of extraneous hemodynamic or toxic triggers, supporting a "multiple hit" hypothesis, in which the accumulation of risk factors (genetic and environmental) increases the likelihood of developing a disease phenotype.

Myocarditis can be triggered by exposure to foreign antigen–most commonly viral– and autoimmunity. In previous studies, *DMD* encoding the protein dystrophin, has been identified as a potential susceptibility gene for myocardial viral infection[32,33] and while only one patient carried a pathogenic variant in *DMD* in this review, it seems likely that variants in other genes associated with cardiomyopathy also heighten susceptibility to myocardial viral infection. With respect to autoimmunity, chronic inflammation may be a determinant of disease progression in some cardiomyopathy phenotypes, most notably DCM, but the interaction between immune effectors and variants in sarcomeric protein genes requires further study.

Less commonly, P/LP variants in non-*TTN* genes, such as those encoding other sarcomeric proteins (e.g., *MYH7*, *MYBPC3*, *TNNI3*, *TNNT2*, and *TNNC1*) or non-sarcomeric protein (e.g., *SCN5A*, *BAG3*) have been identified in patients with complicated myocarditis. Some of these variants have previously been associated with hypertrophic cardiomyopathy. In patients carrying these gene variants, inflammation may play a role in triggering ventricular arrhythmias, promoting the progression of myocardial fibrosis, and influencing adverse events[29, 34, 35].

Age and genetic variants

Alongside clinical presentation, we observed a correlation between the age at onset of myocarditis, its severity, and specific genes affected by P/LP variants. Patients with complicated myocarditis were notably younger than those with uncomplicated myocarditis[7, 9], and the prevalence of P/LP variants in complicated myocarditis was significantly higher in children compared to adults (45% vs. 22%). In adults, variants in *TTN* and in desmosomal genes were more prevalent than in children, aligning with previous data indicating an age-dependent risk of disease occurrence[36, 37]. Of clinical interest, the genes associated with cardiomyopathy in patients with complicated myocarditis overlap with well-known genes responsible for DCM in both adults and children, with *TTN* truncating variants representing the most common in adults[31] and non-*TTN* sarcomeric genes variants (e.g., *MYH7*, *MYPBC3*, *TNNI3*, and *TNNT2*) the most prevalent in children[38].

Clinical implications

If representative of real-world experience, the findings in this review have important implications for clinical practice. Patients with myocarditis and P/LP variants have been found to have worse outcomes compared to those without[7, 9, 16] and identification of a P/LP variant might enable prediction of evolution to cardiomyopathy. Genetic screening in myocarditis cases also enables predictive testing in family members who may be similarly

predisposed to disease.

Study limitations

This study had several limitations. First, the available studies were observational or case series with intrinsic methodological and publication biases and only included patients from Europe and the USA, restricting the generalizability of the results to other countries[39]. Second, there was considerable heterogeneity among studies explained in part by small numbers of patients, inclusion of patients with clinically suspected and biopsy-proven myocarditis, and variable definition of complicated and uncomplicated myocarditis in each study. While, in some cases, it was possible to accurately discriminate between patients fulfilling the definition for complicated and uncomplicated myocarditis, in others, the dichotomization was performed according to the general clinical characteristics of the cohort. For example, in the paper by Lota et al[7], we classified patients included in the London cohort as having uncomplicated myocarditis, as they showed higher LVEF and less severe symptoms. Conversely, patients included in the Maastricht cohort, exhibiting low LVEF and more severe symptoms, were classified as having complicated myocarditis. However, this dichotomization did not prevent a possible misclassification of patients within the two groups.

Third, the prevalence of positive genetic testing among patients with uncomplicated myocarditis may also be influenced by the limited use of genetic testing in this context. Finally, the estimation of the diagnostic yield of genetic testing in this subgroup heavily relies on one study[7], emphasizing the need for future research.

Conclusions

This systematic review and meta-analysis reports a high prevalence of P/LP variants in cardiomyopathy-associated genes in patients with myocarditis. The prevalence of P/LP variants and the affected genes vary according to the clinical presentation.

Perspectives

Competency in Medical Knowledge: Genetic predisposition may have a major role in myocarditis, with putatively deleterious variants in genes encoding for cardiomyopathy structure and function detected in large number of cases.

Competency in Patient Care: Genetic testing should be considered in patients with acute myocarditis to guide management, predict outcomes, and identify family members at risk.

Translational Outlook: Human and animal models exploring the mechanisms of acute myocarditis in patients with monogenic variants may enhance prognostic models and identify novel therapeutic targets.

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Figure legends

Figure 1. *PRISMA flow diagram.*

Study selection process.

Figure 2. *Genetic architecture of myocarditis.*

Distribution of genetic variants among cardiomyopathy-associated genes identified in patients with acute myocarditis.

Figure 3. *Prevalence of genetic variants in myocarditis.*

Prevalence of P/LP variants in cardiomyopathy-associated genes in patients with A. uncomplicated myocarditis, B. complicated myocarditis presenting in adulthood, C. complicated myocarditis presenting in childhood.

Abbreviations: LVEF, left ventricular ejection fraction; SVT, sustained ventricular tachycardia; VF, ventricular fibrillation.

Figure 4. *Genetic architecture of myocarditis according to clinical presentation.*

Distribution of genetic variants among cardiomyopathy-associated genes identified in patients with A. uncomplicated myocarditis, B. complicated myocarditis presenting in adulthood, C. complicated myocarditis presenting in childhood.

Central illustration. *Prevalence of P/LP variants in cardiomyopathy-associated genes in patients with myocarditis.*

Among 732 articles identified through the initial database search, 8 met the inclusion criteria, providing data for 586 patients with acute myocarditis (A). A total of 89 P/LP variants in cardiomyopathy-associated genes were identified in 85 patients, with desmosomal and sarcomeric representing the most commonly involved genes (B). The prevalence of P/LP variants varied according to the clinical presentation (C).

Table 1. The table shows details of the studies included in the meta-analysis.
Abbreviations: DCM, dilated cardiomyopathy; HF, heart failure; HNDCM, hypokinetic non-dilated cardiomyopathy; IQR, interquartile range; LVEF, left ventricular ejection fraction; SCD, sudden cardiac death; VAs, ventricular arrhythmias; UK, United Kingdom; USA, United States of America.

First Author	Year	Country	Diagnosis	Inclusion Criteria	Classification	Population
Artico et al	2020	Italy	Biopsy Proven	Refractory Unexplained HF and LV Dysfunction, Unexplained Life-Threatening or Iterative VAs, or Relapsing Myocarditis and Persistent Troponin Increase	Complicated	Adults
Brown et al	2019	USA	Clinically Suspected	Acute HF with reduced cardiac output	Complicated	Children
Kontorovich et al	2021	USA	Clinically Suspected or Biopsy Proven	No Specific Inclusion Criteria	Complicated: LVEF <40% Uncomplicated: LVEF ≥40%	Adults and Children
Lota et al	2022	UK	Clinically Suspected or Biopsy Proven	No Specific Inclusion Criteria Two cohort described: London Cohort (Median LVEF 63% [IQR 57-67]; Maastricht Cohort (Median	Complicated: Patients Enrolled in Maastricht Cohort Uncomplicated: Patients Enrolled in London Cohort	Adults

				LVEF 36% [IQR 24-45])		
Seidel et al	2021	Germany	Biopsy Proven	No Specific Inclusion Criteria	Complicated: DCM Phenotype Uncomplicated: Non-DCM Phenotype Definition of DCM: Presence of LV Dilatation and Systolic Dysfunction	Children
Seidel et al	2022	Germany	Biopsy Proven	Acute Heart Failure with DCM Phenotype	Complicated	Children
Tiron et al	2022	Spain	Clinically Suspected or Biopsy Proven	No Specific Inclusion Criteria	Complicated: LVEF <30%, Cardiogenic Shock or Sustained VAs Uncomplicated: None of the Complicated Criteria	Adults
van der Meulen et al	2022	the Netherlands	Clinically Suspected or Biopsy Proven	Presentation with DCM Phenotype	Complicated	Children

Table 2. Clinical and genetic characteristics of populations described in studies included in the meta-analysis.
*Two different cohorts were analysed in paper by Lota et at. Lon refers to London cohort and Maas to Maastricht cohort.
Abbreviations: ARVC, arrhythmogenic right ventricular cardiomyopathy; IQR, interquartile range; LVEF, left ventricular ejection fraction; NR, not reported.

First Author	Year	Population	Age	Males	Positive Genetic Testing	Genetic Panel	Genetic Variants
Artico et al	2020	36	46 ±15	22 (61)	11 (31)	23 Cardiomyopathy	*TTN* (n = 8); *DSP* (n = 1); *FLNC* (n = 1); *RBM20* (n = 1)

						-Associated Genes	
Brown et al	2019	8	11.5 ±2.5	1 (12)	5 (62)	51-81 Cardiomyopathy -Associated Genes	*TTN* (n = 2); *MYBPC3* (n = 1); *TNNT2* (n = 1); *SCN5A* (n = 1)
Kontorovich et al	2021	117; LVEF available: 83	NR	NR	19 (16); LVEF <40%: 12/55 (22) LVEF >40%: 2/28 (7)	93 Cardiomyopathy -Associated Genes	*PRDM16* (n = 1); *DSP* (n = 3); *DNM2* (n = 1); *DMD* (n = 1); *TTN* (n = 7); *RYR1* (n = 1); *DYSF* (n = 2); *PKP2* (n = 1); *SGCG* (n = 1); *MYH7* (n = 1); *FLNC* (n = 1); *TRDN* (n = 1); *TNNT1* (n = 1)
Lota et al*	2022	336; Lon: 230 Maa: 106	Lon: 33 (IQR 25-45) Maas: 54 (44-54)	Lon: 193 (57) Maas: 65 (61)	27 (8); Lon: 10 (5%) Maas: 17 (16%)	Lon: 169 Cardiomyopathy -Associated Genes Maas: 47 Cardiomyopathy -Associated Genes	*BAG3* (n = 2); *DES* (n = 1); *DSG2* (n= 1); *DSP* (n = 4); *PKP2* (n = 2); *TTN* (n = 9); *LMNA* (n = 3); *RBM20* (n = 2); *TNNC1* (n = 1); *TNNT2* (n = 3)
Seidel et al	2021	42; DCM: 20 Non-DCM: 22	10 (range1.1-16.4); DCM: 1.4 (range 0.3-4.1) Non-DCM: 16.1 (11.5-17.1)	25 (60); DCM: 11 (55) Non-DCM: 14 (64)	9 (21) DCM: 7 (35) Non-DCM: 2 (9)	89 Cardiomyopathy -Associated Genes	*BAG3* (n = 2); *DSP* (n = 2); *LMNA* (n = 1); *MYH7* (n = 1); *TNNI3* (n = 1); *TNNT2* (n = 1); *TTN* (n = 1)
Seidel et al	2022	12	1.6 (range 0.8-8)	5 (42)	8 (67)	89 Cardiomyopathy -Associated Genes	*TTN* (n = 2); *TNNC1* (n = 1); *TNNI3* (n = 2); *MYH7* (n = 2); *RYR2* (n = 1)

Tiron et al	2022	28; Complicated: 12 Uncomplicated: 16	Complicated: 41 (range 25-74); Uncomplicated: 35 (range 18-68)	22 (79); Complicated: 7 (58); Uncomplicated: 15 (94)	5 (18); Complicated: 4 (33) Uncomplicated: 1 (6)	71 Cardiomyopathy-Associated Genes	*FLNC* (n = 1); *RBM20* (n = 1); *BAG3* (n = 2); *DSP* (n= 1)
van der Meulen et al	2022	7	NR	NR	1 (14)	28 or 70 Cardiomyopathy-Associated Genes	*LMNA* (n = 1)

A

Articles Identified from PubMed and Embase
(n = 732)

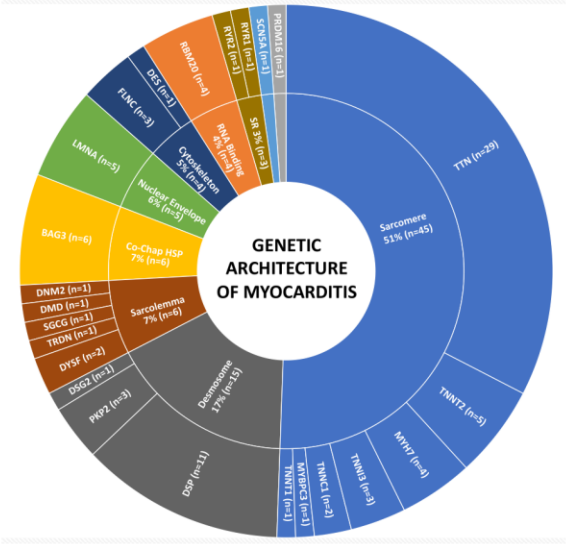
Articles Fulfilling Inclusion Criteria
(n = 8)

Patients with Acute Myocarditis
(n = 586)

P/LP Variants Detected
(n = 89 in 85 patients)

B

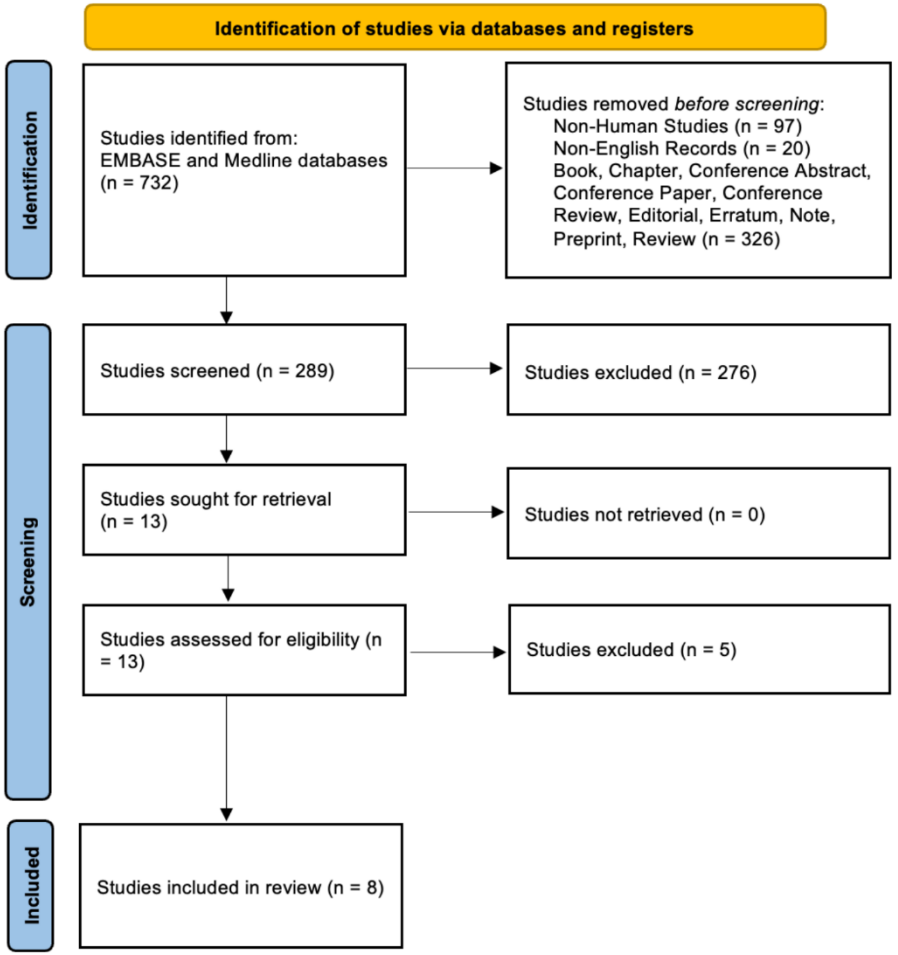
Desmosomal and Sarcomeric Are The Most Common Genes Involved

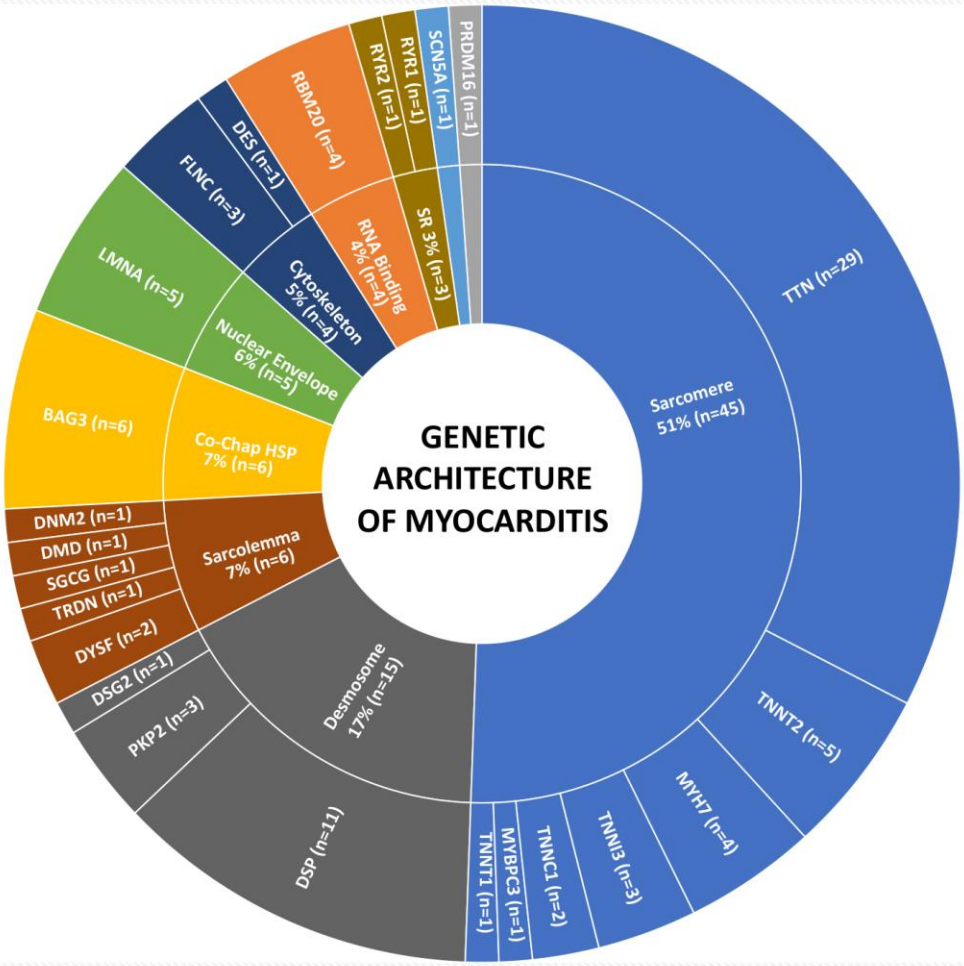


C

Prevalence of P/LP Variants in Cardiomyopathy Associated Genes

4%	22%	44%
Uncomplicated Myocarditis	Complicated Myocarditis (Adults)	Complicated Myocarditis (Children)

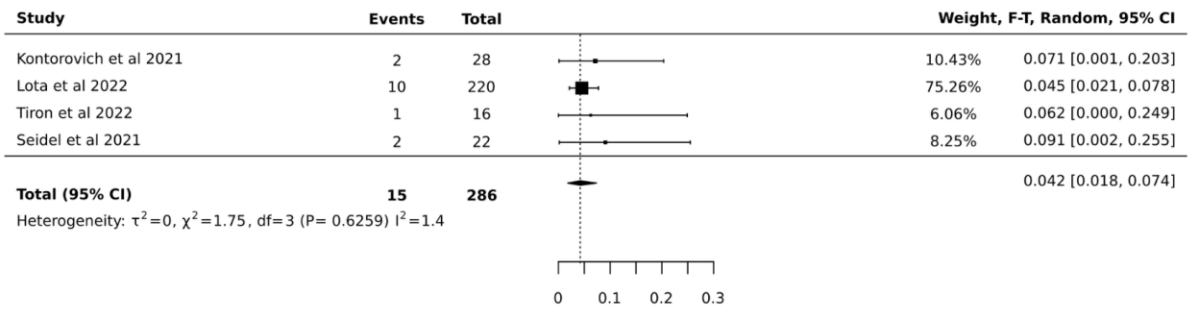

Identification of studies via databases and registers
Identification
Studies identified from:
EMBASE and Medline databases
(n = 732)
Studies removed *before screening*:
Non-Human Studies (n = 97)
Non-English Records (n = 20)
Book, Chapter, Conference Abstract, Conference Paper, Conference Review, Editorial, Erratum, Note, Preprint, Review (n = 326)
Screening
Studies screened (n = 289)
Studies excluded (n = 276)
Studies sought for retrieval
(n = 13)
Studies not retrieved (n = 0)
Studies assessed for eligibility (n = 13)
Studies excluded (n = 5)
Included
Studies included in review (n = 8)


GENETIC ARCHITECTURE OF MYOCARDITIS
Sarcomere 51% (n=45)
TTN (n=29)
TNNT2 (n=5)
MYH7 (n=4)
TNNI3 (n=3)
TNNC1 (n=2)
MYBPC3 (n=1)
TNNT1 (n=1)
Desmosome 17% (n=15)
DSP (n=11)
PKP2 (n=3)
DSG2 (n=1)
Sarcolemma 7% (n=6)
DYSF (n=2)
TRDN (n=1)
SGCG (n=1)
DMD (n=1)
DNM2 (n=1)
Co-Chap HSP 7% (n=6)
BAG3 (n=6)
Nuclear Envelope 6% (n=5)
LMNA (n=5)
Cytoskeleton 5% (n=4)
FLNC (n=3)
DES (n=1)
RNA Binding 4% (n=4)
RBM20 (n=4)
SR 3% (n=3)
RYR2 (n=1)
RYR1 (n=1)
SCN5A (n=1)
PRDM16 (n=1)

A

Uncomplicated Myocarditis

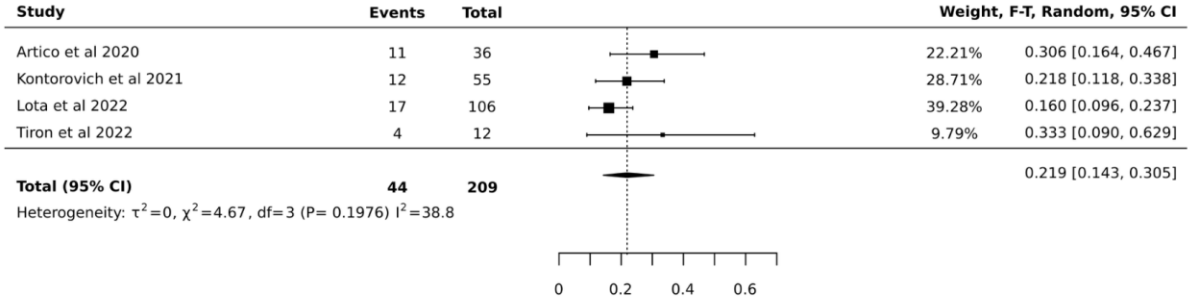
Definition: Normal LVEF, No SVT/VF



B

Complicated Myocarditis Presenting in Adulthood

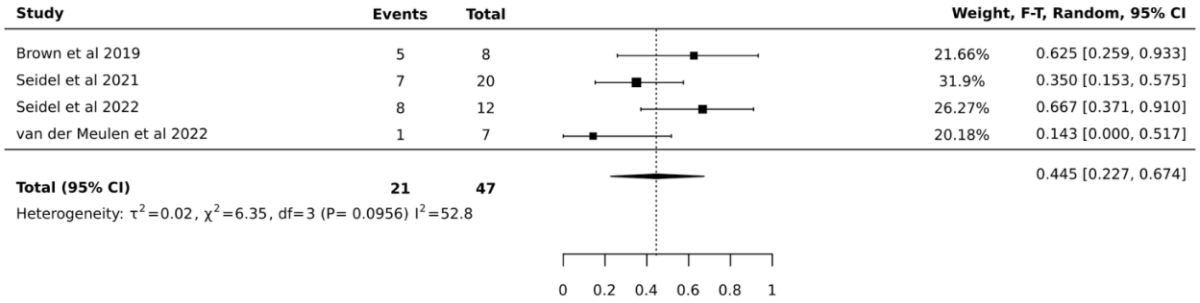
Definition: Reduced LVEF and/or SVT/VF

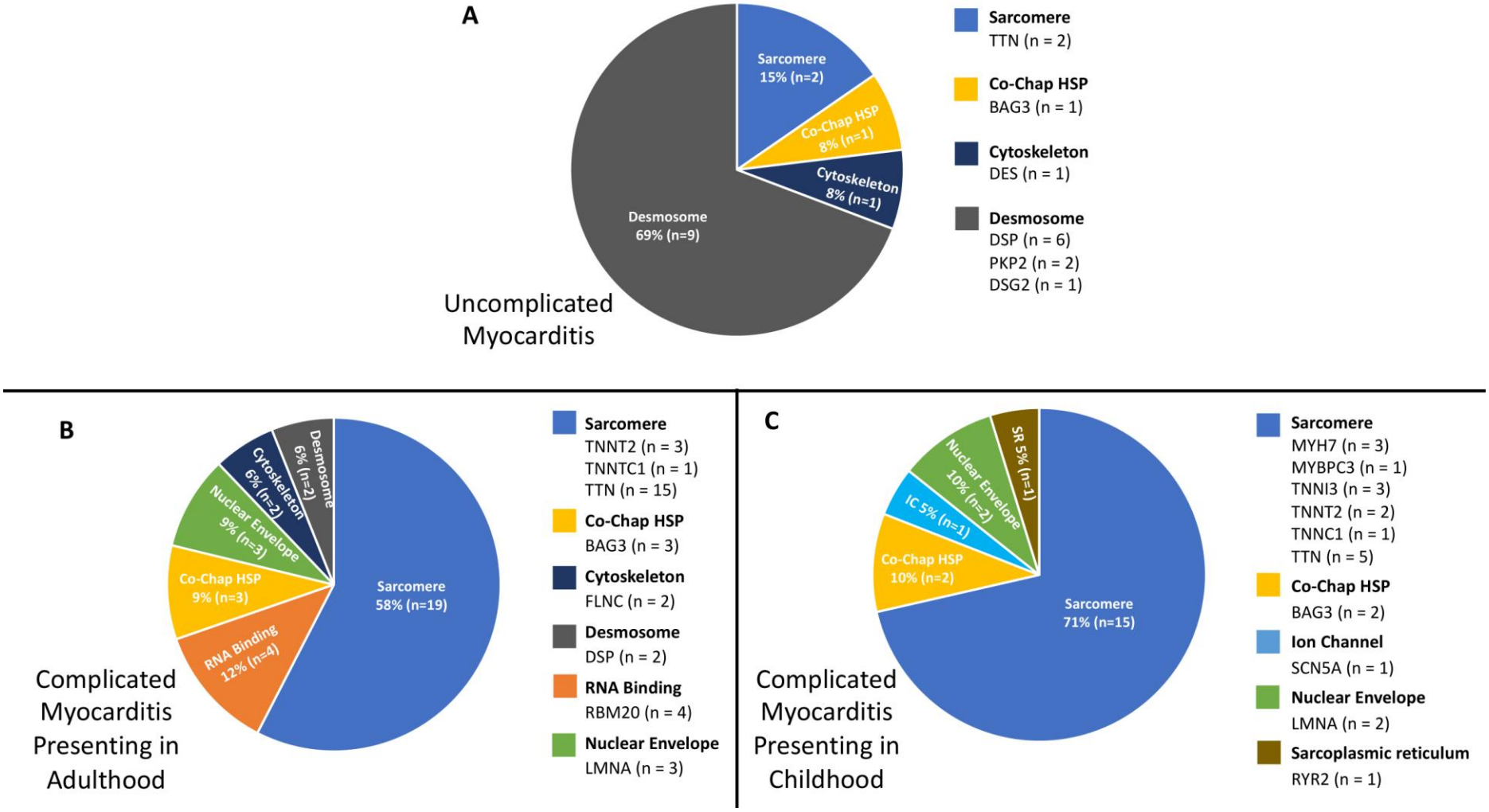


C

Complicated Myocarditis Presenting in Childhood

Definition: Reduced LVEF and/or SVT/VF




A
Sarcomere 15% (n=2)
Co-Chap HSP 8% (n=1)
Cytoskeleton 8% (n=1)
Desmosome 69% (n=9)
Uncomplicated Myocarditis
Sarcomere
TTN (n = 2)
Co-Chap HSP
BAG3 (n = 1)
Cytoskeleton
DES (n = 1)
Desmosome
DSP (n = 6)
PKP2 (n = 2)
DSG2 (n = 1)
B
Sarcomere 58% (n=19)
RNA Binding 12% (n=4)
Co-Chap HSP 9% (n=3)
Nuclear Envelope 9% (n=3)
Cytoskeleton 6% (n=2)
Desmosome 6% (n=2)
Complicated Myocarditis Presenting in Adulthood
Sarcomere
TNNT2 (n = 3)
TNNTC1 (n = 1)
TTN (n = 15)
Co-Chap HSP
BAG3 (n = 3)
Cytoskeleton
FLNC (n = 2)
Desmosome
DSP (n = 2)
RNA Binding
RBM20 (n = 4)
Nuclear Envelope
LMNA (n = 3)
C
Sarcomere 71% (n=15)
Co-Chap HSP 10% (n=2)
IC 5% (n=1)
Nuclear Envelope 10% (n=2)
SR 5% (n=1)
Complicated Myocarditis Presenting in Childhood
Sarcomere
MYH7 (n = 3)
MYBPC3 (n = 1)
TNNI3 (n = 3)
TNNT2 (n = 2)
TNNC1 (n = 1)
TTN (n = 5)
Co-Chap HSP
BAG3 (n = 2)
Ion Channel
SCN5A (n = 1)
Nuclear Envelope
LMNA (n = 2)
Sarcoplasmic reticulum
RYR2 (n = 1)

Supplemental Table 1. MOOSE checklist for meta-analysis of observational studies.

Reporting Criteria	Reported	Reported on Page No.
Reporting of Background		
Problem definition	Yes	4
Hypothesis statement	Yes	4
Description of Study Outcome(s)	Yes	6
Type of exposure or intervention used	Yes	4-6
Type of study design used	Yes	5
Study population	Yes	5-7
Reporting of Search Strategy		
Qualifications of searchers (eg, librarians and investigators)	Yes	5
Search strategy, including time period included in the synthesis and keywords	Yes	5
Effort to include all available studies, including contact with authors	N/A	-
Databases and registries searched	Yes	
Search software used, name and version, including special features used (eg, explosion)	Yes	5
Use of hand searching (eg, reference lists of obtained articles)	Yes	5
List of citations located and those excluded, including justification	Yes	5
Method for addressing articles published in languages other than English	N/A	-
Method of handling abstracts and unpublished studies	Yes	5
Description of any contact with authors	N/A	-
Reporting of Methods		
Description of relevance or appropriateness of studies assembled for assessing the hypothesis to be tested	Yes	5-6

Rationale for the selection and coding of data (eg, sound clinical principles or convenience)	Yes	5-6
Documentation of how data were classified and coded (eg, multiple raters, blinding, and interrater reliability)	Yes	5-6
Assessment of confounding (eg, comparability of cases and controls in studies where appropriate)	Yes	6
Assessment of study quality, including blinding of quality assessors; stratification or regression on possible predictors of study results	Yes	6
Assessment of heterogeneity	Yes	6
Description of statistical methods (eg, complete description of fixed or random effects models, justification of whether the chosen models account for predictors of study results, dose-response models, or cumulative meta-analysis) in sufficient detail to be replicated	Yes	6
Provision of appropriate tables and graphics	Yes	17-21, Supplemental Material
Reporting of Results		
Table giving descriptive information for each study included	Yes	18-21
Results of sensitivity testing (eg, subgroup analysis)	Yes	Figures and Supplemental Material
Indication of statistical uncertainty of findings	N/A	-
Reporting of Discussion		
Quantitative assessment of bias (eg, publication bias)	Yes	10-13
Justification for exclusion (eg, exclusion of non–English-language citations)	N/A	-
Assessment of quality of included studies	Yes	10-13
Reporting of Conclusions		
Consideration of alternative explanations for observed results	Yes	12-13

Generalization of the conclusions (ie, appropriate for the data presented and within the domain of the literature review)	Yes	10-13
Guidelines for future research	N/A	-
Disclosure of funding source	Yes	1

Supplemental Table 2. The table displays for each database consulted its name, the platform through which the databases was searched, the query used for the search and the results.

Database	Query	Results
Embase Embase Pubmed	('myocarditis'/exp OR 'myocarditis') AND ('genetic testing'/exp OR 'genetic testing' OR 'genetic test' OR 'genetic tested' OR 'genetic analysis'/exp OR 'genetic analysis' OR 'genetic screening'/exp OR 'genetic screening' OR 'genetic screen' OR 'molecular test'/exp OR 'molecular test' OR 'molecular testing'/exp OR 'molecular testing' OR 'molecular analysis'/exp OR 'molecular analysis')	732

Supplemental Table 3. Definition of acute myocarditis as reported in the studies included in the meta-analysis.

First Author	Study Population and Inclusion Criteria
Artico et al. 2020	This study included **adults** (n = 36; age 46 ±15 years) with **biopsy-proven** active lymphocytic myocarditis according to Dallas criteria and immunohistochemical analysis. Indications for biopsy were refractory unexplained heart failure and left ventricular systolic (75%, n = 27), unexplained life-threatening or iterative ventricular arrhythmias (17%, N = 6), or relapsing myocarditis and persistent troponin increase despite normal left ventricular ejection fraction (8%, n = 3).
Brown et al. 2019	This study included **children** (n = 8; age 11.5 ±2.5 years) who presented in the paediatric intensive care unit with acute heart failure tentatively suspected to be acute myocarditis. The individuals were presumed to have myocarditis due to the acute onset, apparent lack of a family history and no significant medical history to suggest another aetiology. All the patients had decreased left ventricular systolic function and all patients had elevated troponin I levels.
Kontorovich et al. 2021	This study included **adults and children** (n = 117; age not reported) with acute myocarditis obtained from three different registries: - AM1: Cases were adults and children with **clinical diagnosis of acute myocarditis** based on historical and diagnostic testing data who were recruited between 1991 and 2000; - AM2: Cases were subjects with clinically suspected viral myocarditis recruited between 2015 and 2017, with acute myocarditis **proven by immunohistologic criteria**; - AM3: Cases were subjects with clinically suspected viral myocarditis recruited between 1991 and 2000, with acute myocarditis **proven by immunohistologic criteria**.
Lota et al. 2022	This study included adults from two different cohort: - London cohort: **Adults** with myocarditis (n = 220; Age 33 [IQR 22-45] years) were evaluated by **cardiovascular magnetic resonance or immunohistopathology of myocardial tissue** by European Society of Cardiology Criteria*. Of these, 114 cases were consecutively recruited <14 days after acute hospitalization, and 116 were retrospectively identified with cardiovascular magnetic resonance or biopsy-confirmed acute myocarditis. Exclusion criteria were coronary artery disease and congenital heart disease. - Maastricht cohort: **Adults** with myocarditis (n = 106; Age 54 [IQR 44-54] years) were **confirmed on endomyocardial biopsy** within six months of acute presentation with suspected myocarditis. Exclusion criteria matched those of the London cohort

Seidel et al. 2021	This study included **children** (n = 42; median age 10 [range 1.1-16.4] years) with acute myocarditis with diagnosis confirmed **according to established histological and immunohistochemical criteria**. Endomyocardial biopsy was performed as the gold standard for the diagnosis of myocarditis, new-onset heart failure or ventricular arrhythmia.
Seidel et al. 2022	This study included **children** (n = 12; median age 1.6 [range 0.8-8.0] years) with **biopsy-proven myocarditis** and dilated cardiomyopathy phenotype at admission.
Tiron et al. 2022	This study included **adults** (n = 28; age range 18-68) with myocarditis diagnosed following the European Society of Cardiology criteria, taking into account clinical presentation, ECG, myocardial cytolysis markers, echocardiography, and tissue characterization by cardiac magnetic resonance imaging or endomyocardial biopsy.
van der Meulen et al. 2022	This study included **children** (n = 7; age not reported) with diagnosis based on histological or immune-histological evidence of myocarditis or based on clinical features and viral test results.

Supplemental Table 4. Definition of complicated and uncomplicated myocarditis in the different studies.

First Author	Definitions
Artico et al. 2020	**Definition of complicated myocarditis:** individuals with refractory unexplained heart failure and left ventricular dysfunction, unexplained life-threatening or iterative ventricular arrhythmias, or relapsing myocarditis and persistent troponin increase despite normal left ventricular ejection fraction.
Brown et al. 2019	**Definition of complicated myocarditis:** individuals with acute-onset heart failure with decreased left ventricular systolic function (with or without left ventricular dilation).
Kontorovich et al. 2021	The myocarditis severity in this study was based on the presence of left ventricular ejection fraction, that was available only for a subset of patients. **Definition of complicated myocarditis:** individuals with left ventricular ejection fraction < 40%. **Definition of uncomplicated myocarditis**: individuals with left ventricular ejection fraction ≥ 40%
Lota et al. 2022	This study included adults from two different cohort: - London cohort, including individuals with a median left ventricular ejection fraction of 63% (IQR 57-67) and prevalence of patients in NYHA class III/IV of 5%. - Maastricht cohort, including individuals with a median left ventricular ejection fraction of 36% (IQR 24-45) and prevalence of patients in NYHA class III/IV of 38%. **Definition of complicated myocarditis:** individuals included in the Maastricht cohort. **Definition of uncomplicated myocarditis:** individuals included in the London cohort.
Seidel et al. 2021	The cohort was divided into two subgroups, according to the presence of dilated cardiomyopathy phenotype. **Definition of complicated myocarditis:** individuals with dilated cardiomyopathy phenotype (left ventricular dilation and dysfunction). **Definition of uncomplicated myocarditis:** individuals without dilated cardiomyopathy phenotype.
Seidel et al. 2022	**Definition of complicated myocarditis:** individuals with dilated cardiomyopathy phenotype (left ventricular dilation and dysfunction).
Tiron et al. 2022	**Definition of complicated myocarditis:** individuals with severe ventricular dysfunction (left ventricular ejection fraction, <30%), cardiogenic shock, or sustained ventricular arrhythmias. **Definition of non-severe myocarditis:** individuals without criteria for severe myocarditis.
van der Meulen et al. 2022	**Definition of complicated myocarditis:** individuals with dilated cardiomyopathy phenotype (left ventricular dilation and dysfunction).

Supplemental Table 5. Pathogenic/likely pathogenic variants identified in the included studies.

First Author	Year	Gene	Reported Variant	Reported Classification	Method of Variant Classification
Artico et al	2020	TTN	Not specified	Pathogenic or Likely Pathogenic variants	Not specified
		TTN	Not specified		
		TTN	Not specified		
		TTN	Not specified		
		TTN	Not specified		
		TTN	Not specified		
		TTN	Not specified		
		TTN	Not specified		
		DSP	Not specified		
		FLNC	Not specified		
		RBM20	Not specified		
Brown et al	2019	TTN	(p.Pro28826fs)	Likely Pathogenic	ACMG criteria
		TTN	(p.Arg21747Ter)	Pathogenic	
		MYBPC3	(p.Glu542Gln)	Pathogenic	
		TNNT2	(p.Lys210del)	Pathogenic	
		SCN5A	(p.Arg1898His)	Likely Pathogenic	
Kontorovich et al	2021	PRDM16	c.420delG	Putatively damaging variants	Classified as putatively deleterious variants if previously interpreted as "likely pathogenic" or "pathogenic" with no conflicting evidence status among Badge laboratories entries on ClinVar.
		DSP	c.3697dupA		
		DSP	c.1234C>T		
		DSP	c.5851C>T		
		DNM2	c.1347dupC		
		DMD	c.823dupT		
		TTN	c.76806dupA		
		TTN	c.52867_52868insCA		
		TTN	c.35680C>T		
		TTN	c.51250delA		
		TTN	c.22552C>T		

		TTN	c.50536delT		
		TTN	c.38004delA		
		TTN	c.38004delA		
		RYR1	c.1589G>A		
		DYSF	c.4152dupC		
		DYSF	c.760C>T		
		PKP2	c.1771delC		
		SGCG	c.581T>C		
		MHY7	c.2377C>T		
		FLNC	c.7870delA		
		TRDN	c.991+2T>A		
		TNNT1	c.73G>T		
Lota et al	2022	BAG3	c.235del (p.Ala79LeufsTer132)	Likely Pathogenic	ACMG criteria
		BAG3	c.910C>T (p.Gln304*)	Likely Pathogenic	
		DES	c.1048C>T (p.Arg350Trp)	Likely Pathogenic	
		DSG2	c.829_840del (p.Leu277_Met280del)	Likely Pathogenic	
		DSP	c.4307_4308del (p.Thr1436ArgfsTer3)	Likely Pathogenic	
		DSP	c.4423del (p.Thr1475ProfsTer9)	Likely Pathogenic	
		DSP	c.5056C>T (p.Q1686X)	Likely Pathogenic	
		DSP	c.6393del (p.Gly2133Valfs*2)	Likely Pathogenic	
		PKP2	c.968_969del (p.Gln323ArgfsTer12)	Likely Pathogenic	
		PKP2	c.337-2A>T	Likely Pathogenic	
		TTN	c.90688G>T (p.G30230X)	Likely Pathogenic	
		TTN	c.51459_51462del (p.Asp17153GlufsTer11)	Likely Pathogenic	

		TTN	c.61921C>T (p.Arg20641*)	Likely Pathogenic	
		TTN	c.13100del (p.Lys4367Argfs*27)	Likely Pathogenic	
		TTN	c.73332C>A (p.Cys24444*)	Likely Pathogenic	
		TTN	c.64688del (p.Pro21563Leufs*10)	Likely Pathogenic	
		TTN	c.87782del (p.Pro29261Glnfs*10)	Likely Pathogenic	
		TTN	c.13100del (p.Lys4367Argfs*27)	Likely Pathogenic	
		TTN	c.65042del (p.Asp21681Alafs*15)	Likely Pathogenic	
		LMNA	c.992G>A (p.Arg331Gln)	Likely Pathogenic	
		LMNA	c.1517A>C (p.His506Pro)	Likely Pathogenic	
		LMNA	c.647G>A (p.Arg216His)	Likely Pathogenic	
		RBM20	c.1900C>T (p.Arg634Trp)	Likely Pathogenic	
		RBM20	c.1764T>G (p.Ile588Met)	Likely Pathogenic	
		TNNC1	c.317+1G>A (p.Gly68Glufs*12)	Likely Pathogenic	
		TNNT2	c.442C>T (p.Arg148Trp)	Likely Pathogenic	
		TNNT2	c.742T>G (p.Phe248Val)	Likely Pathogenic	
		TNNT2	c.416G>A (p.Arg139His)	Likely Pathogenic	
Seidel et al	2021	BAG3	c.608delG (p.Tyr205Thrfs*6)	Pathogenic (PM2, PVS1)	ACMG criteria
		BAG3	c.925C>T (p.Arg309*)	Pathogenic (PM2, PVS1, PM6)	

		DSP	c.2200A>del (p.Arg734Glufs*31)	Likely Pathogenic (PM2, PVS1)	
		DSP	c.4372C>T (p.Arg1458*)	Likely Pathogenic (PM2, PVS1)	
		LMNA	c.868G>A (p.Glu290Lys)	Pathogenic (PM1, PM2, PS1, PP3)	
		MYH7	c.644C>T (p.Thr215Ile)	Likely Pathogenic (PM1-2, PM6, PP3)	
		TNNI3	c.204delG (p.Arg68Argfs*9)	Pathogenic (PM2, PS3, PVS1)	
		TNNT2	c.460C>T (p.Arg154Trp)	Likely Pathogenic (PM2, PS1, PP3)	
		TTN	c.25889_25892del (p.E8630Gfs*28)	Likely Pathogenic (PM1-2, PM4)	
Seidel et al	2022	TTN	c.66547C>T (p.R22183*)	Likely Pathogenic	ACMG criteria
		TTN	c.24597C>A (p.Y8199*)	Likely Pathogenic	
		TNNC1	c.100G>A (p.G34S)	Likely Pathogenic	
		TNNI3	c.146T>A (p.L49Q)	Likely Pathogenic	
		TNNI3	c.544G>C (p.E182Q)	Likely Pathogenic	
		MYH7	c.644C>T (p.T215I)	Likely Pathogenic	
		MYH7	c.1633G>A (p.D545N	Likely Pathogenic	
		RYR2	c.3265G>A (p.E1089K)	Likely Pathogenic	
Tiron et al	2022	FLNC	p.Pro1555Leufs*52	Pathogenic or Likely Pathogenic variants	ACMG criteria
		RBM20	c.1104_1585+467del		

		BAG3	p.Gln88*		
		BAG3	p.Gln88*		
		DSP	p.Gln113*		
van der Meulen et al	2022	LMNA	c.992G>A (p.Arg331Gln)	Pathogenic Variant (Class 5)	ACMG criteria

Supplemental Table 6. Critical appraisal of eligible studies and risk of bias.

Citation	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Risk of Bias
Artico et al. 2020	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	N/A	High
Brown et al. 2019.	No	No	No	Yes	Yes	Yes	No	Yes	N/A	High
Kontorovich et al. 2021	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	N/A	Low
Lota et al. 2022	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	N/A	Low
Seidel et al. 2021	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	N/A	High
Seidel et al. 2022.	No	No	No	Yes	No	Yes	Yes	Yes	N/A	High
Tiron et al. 2022	No	No	No	Yes	Yes	Yes	Yes	Yes	N/A	High
Van Der Meulen et al. 2022	No	No	No	Yes	Yes	Yes	Yes	Yes	N/A	High

Definitions: **Q1:** Was the sample frame appropriate to address the target population? **Q2:** Were study participants recruited in an appropriate way? **Q3:** Was the sample size adequate? **Q4:** Were the study subjects and setting described in detail? **Q5:** Was data analysis conducted with sufficient coverage of the identified sample? **Q6:** Were valid methods used for the identification of the condition? **Q7:** Was the condition measured in a standard, reliable way for all participants? **Q8:** Was there appropriate statistical analysis? **Q9:** Was the response rate adequate, and if not, was the low response rate managed appropriately?

Supplemental Table 7. STROBE checklist for observational studies included in the systematic review and meta-analysis.
Abbreviations: NR, not reported.

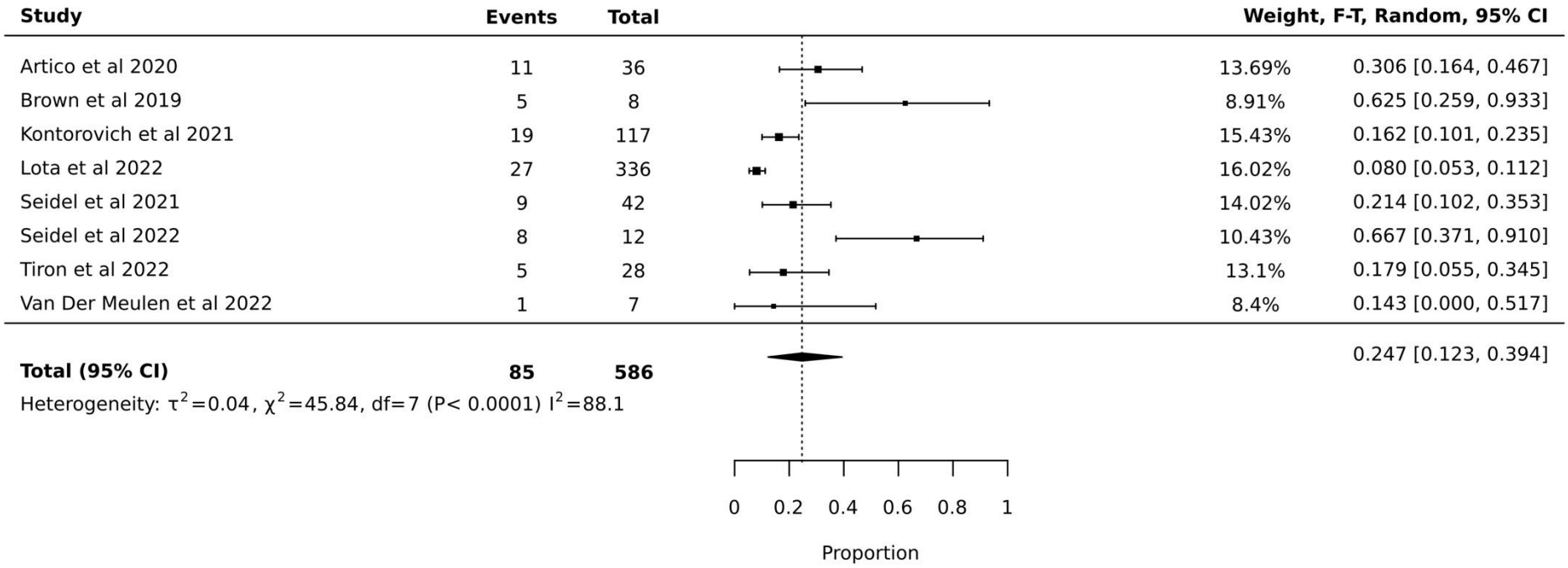
Reporting Criteria	Recommendation	Artico et al. 2020	Brown et al. 2019.	Kontorovich et al. 2021	Lota et al. 2022	Seidel et al. 2021	Seidel et al. 2022.	Tiron et al. 2022	Van Der Meulen et al. 2022
Title and Abstract	Indicate the study's design with a commonly used term in the title or the abstract	NR	Yes	Yes	Yes	Yes	Yes	NR	Yes
	Provide in the abstract an informative and balanced summary of what was done and what was found	NR	Yes	Yes	Yes	Yes	Yes	NR	Yes
Introduction									
Background/rationale	Explain the scientific background and rationale for the investigation being reported	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Objectives	State specific objectives, including any prespecified hypotheses	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Methods									
Study design	Present key elements of study design early in the paper	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Setting	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection	NR	Yes	Yes	Yes	Yes	Yes	NR	Yes
Participants	Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up	Yes	Yes	Yes	Yes	Yes	Yes	NR	Yes

Variables	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable	NR	Yes	Yes	Yes	Yes	Yes	NR	Yes
Data sources/ measurement	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group	NR	Yes	Yes	Yes	Yes	Yes	NR	Yes
Bias	Describe any efforts to address potential sources of bias	NR	NR	Yes	Yes	Yes	NR	NR	Yes
Study size	Explain how the study size was arrived at	NR	Yes	Yes	Yes	Yes	Yes	NR	Yes
Quantitative variables	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why	NR	NR	Yes	Yes	Yes	Yes	NR	Yes
Statistical methods	Describe all statistical methods, including those used to control for confounding	NR	NR	Yes	Yes	Yes	Yes	NR	Yes
	Describe any methods used to examine subgroups and interactions	NR	NR	Yes	Yes	Yes	Yes	NR	Yes
	Explain how missing data were addressed	NR	NR	Yes	Yes	Yes	Yes	NR	Yes
	If applicable, explain how loss to follow-up was addressed	NR	NR	NR	Yes	Yes	NR	NR	Yes
	Describe any sensitivity analyses	NR	NR	Yes	Yes	Yes	Yes	NR	Yes
Results									

Participants	Report numbers of individuals at each stage of study—eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
	Give reasons for non-participation at each stage	NR	NR	Yes	Yes	Yes	NR	NR	Yes
	Consider use of a flow diagram	NR	NR	NR	NR	Yes	NR	NR	NR
Descriptive data	Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
	Indicate number of participants with missing data for each variable of interest	NR	NR	NR	Yes	Yes	Yes	NR	NR
	Summarise follow-up time (eg, average and total amount)	Yes	Yes	Yes	Yes	Yes	Yes	NR	Yes
Outcome data	Report numbers of outcome events or summary measures over time	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Main results	Give unadjusted estimates and, if applicable, confounder-adjusted estimates and their precision (eg, 95% confidence interval). Make clear which confounders were adjusted for and why they were included	NR	NR	Yes	Yes	Yes	Yes	NR	Yes
	Report category boundaries when continuous variables were categorized	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

	If relevant, consider translating estimates of relative risk into absolute risk for a meaningful time period	NR	NR	NR	NR	NR	NR	NR	NR
Other analyses	Report other analyses done—eg analyses of subgroups and interactions, and sensitivity analyses	Yes	NR	Yes	Yes	Yes	Yes	NR	Yes
Discussion									
Key results	Summarise key results with reference to study objectives	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Limitations	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Interpretation	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Generalisability	Discuss the generalisability (external validity) of the study results	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Other information									
Funding	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

Supplemental Figure 1. Pooled prevalence of P/LP variants in cardiomyopathy-associated genes in the overall population.



Supplemental Figure 2. Pooled prevalence of P/LP variants in cardiomyopathy-associated genes in the adults, regardless of clinical presentation.