Kelvin Open Science Publishers
Connect with Research Community

Review Article Volume 1 | Issue 2

KOS Journal of Public Health and Integrated Medicine
https://kelvinpublishers.com/journals/public-health-and-integrated-medicine.php

Novel Therapies for Transthyretin Amyloid Cardiomyopathy

Claudia Gabriele Werner* **and Frank-Dietrich Wagner**

Charité Research Organisation, Berlin, Germany

***Corresponding author:** Claudia Gabriele Werner, Charité Research Organisation, Berlin, Germany

Received: July 30, 2025; **Accepted:** August 09, 2025; **Published:** August 15, 2025

Citation: Claudia GW, et al. (2025) Novel Therapies for Transthyretin Amyloid Cardiomyopathy. *KOS J Pub Health Int Med.* 1(2): 1-15.

1. Abstract

Cardiac amyloidosis caused by transthyretin is a rare but severe cardiomyopathy caused by aberrant accumulation of misfolded transthyretin protein (TTR) in the myocardium. If untreated, transthyretin amyloid cardiomyopathy (ATTR-CM) is characterized by progressive worsening heart failure with a median survival of less than 4 years. Complications include cardiac arrhythmias and conduction system diseases, which can cause sudden cardiac death due to fatal arrhythmias or complete heart block. Functional capacity and quality of life deteriorate exponentially with each worsening of heart failure and subsequent hospitalization.

ATTR-CM was underdiagnosed in the past as its symptoms may overlap with those of other cardiac diseases and efficient diagnostic technologies were missing. Thanks to the availability of non-invasive scintigraphy-based imaging, early diagnosis is now increasingly possible without the need for cardiac biopsy.

Current approved TTR-specific therapies either block the formation of mutated TTR protein by mRNA silencing (vutrisiran) or stabilize the TTR tetramer to prevent misfolded transthyretin protein (tafamidis). The next generation TTR stabilizer acoramidis has recently been approved for the treatment of ATTR-CM due to its success in the ATTRibute-CM trial. Novel mRNA silencer agents are currently investigated in large multicenter phase 3 trials such as eplontersen (CARDIO-TTRansform, NCT04136171) and nucresiran (TRITON-CM, NCT07052903). Inhibition of TTR synthesis is currently also approached with novel gene-editing agents including nexiguran-ziclumeran and YOLT-201.Novel strategies include ATTR amyloid depletion by monoclonal antibodies (ALXN2220, AT-02 or coramitug) and inhibition of TTR fibril formation by the seeding inhibitor TabFH2 or molecular tweezer CLR01.

2. Keywords

Transthyretin amyloid cardiomyopathy, TTR stabilizer, TTR gene silencer, TTR gene-editing, ATTR amyloid depleter

3. Introduction

3.1. Subtypes of cardiac amyloidosis

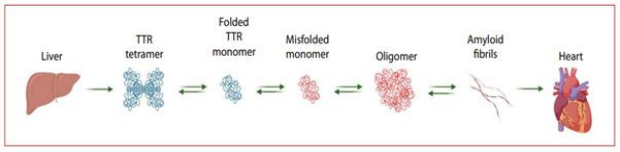
Amyloidosis is a systemic disease in which amyloid fibrils are deposited locally or throughout the body due to the misfolding of unstable amyloid proteins. The vast majority of amyloidosis is caused by one of two proteins: Light chain amyloidosis (AL) or transthyretin amyloidosis (ATTR). The deposition of amyloid fibrils in the extracellular spaces of the heart tissue can cause cardiomyopathy (AL-CM or ATTR-CM) that is mainly characterized by a diastolic form of heart failure and cardiac arrhythmias.

AL-CM can be caused by malignant bone marrow diseases such as multiple myeloma, where malignant plasma cell

clones can produce immunoglobulin light-chains in excess that are prone to misfold into beta-pleated sheets (AL aggregates). Treatment of AL-CM consists mainly of modern chemotherapies that can break down these AL aggregates. Management of AL-CM by a team of blood cancer specialists and cardiologists is crucial for a successful therapy.

ATTR-CM is a progressive worsening disease caused by the extracellular deposition of misfolded transthyretin protein in the heart. Transthyretin is an abundant protein produced by the liver and functions as a transporter of thyroxine and vitamin A (retinol). It normally circulates predominantly as a homotetramer, with a small amount of transthyretin circulating in monomeric form. The monomeric form of transthyretin is prone to misfold and is gradually deposited as amyloid fibrils in the heart causing organ dysfunction [1] (Figure 1).

Figure 1: Transthyretin amyloid formation.

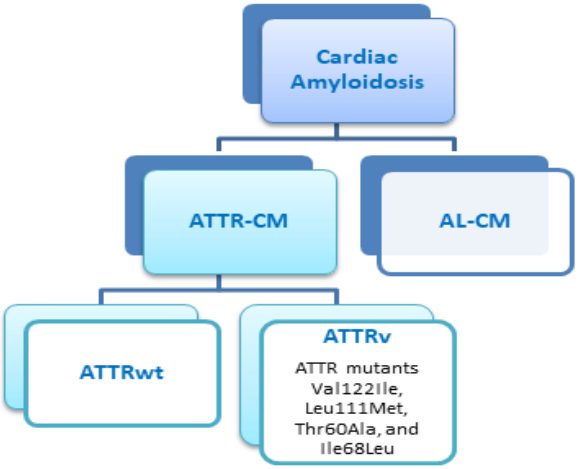


Transthyretin is mainly synthesized in the liver and it circulates predominantly as a homotetramer, with a small amount of transthyretin circulating in monomeric form. The monomeric form of transthyretin is prone to misfold and to self-assemble into oligomers and amyloid fibrils. Transthyretin amyloid fibrils gradually deposit in the heart causing organ dysfunction.

Source: Figure adopted from Wu & Chen, 2024 [1].

Based on whether TTR has a gene mutation, two main subtypes of ATTR amyloidosis exist: wild-type ATTR (ATTRwt), and mutant variants of ATTR (ATTRv) with autosomal dominant heredity (Figure 2). The gene coding for the TTR protein is present on chromosome 18. So far, more than 150 gene mutations have been identified to cause ATTRv. The most common mutation Val30Met is associated primarily with neuropathy (ATTR-PN) and is endemic in Portugal, Japan, Sweden, and Majorca [2,3]. Other mutations, namely Val122Ile, Leu111Met, Thr60Ala, and Ile68Leu, represent the major causative variants associated with cardiomyopathy (ATTR-CM). Val122Ile is primarily prevalent among individuals of African American descent while Thr60Ala is more frequently observed in individuals of Irish heritage. The remaining two mutations are more commonly encountered in populations from Denmark and Italy [4].

Figure 2: Subtypes of cardiac amyloidosis.

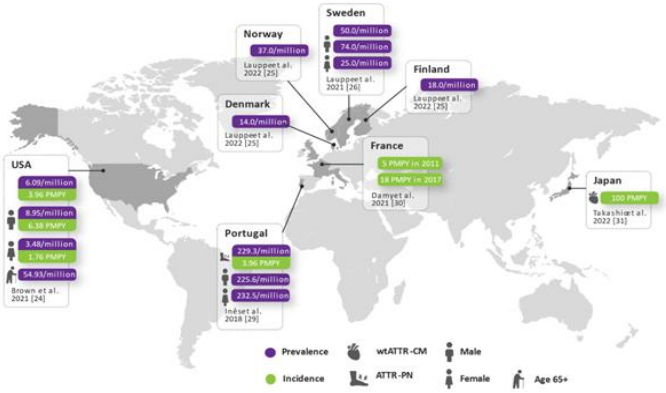


In the case of ATTRwt, the transthyretin protein is the normal (nonmutated) TTR tetramer protein that easily dissociates into misfolding monomers, which over a period of decades gradually deposit as amyloid fibrils. Although small amounts of amyloid deposits can occur in the soft tissue (causing carpal tunnel syndrome) and vasculature, the primary pathologic deposits occur in the heart muscle resulting in cardiac dysfunction (ATTR-CM). Clinical manifestations of the two major phenotypes, ATTR-PN and ATTR-CM often overlap depending on the mutation variant, age, gender and other factors [5] (Appendix 1).

3.2. Worldwide prevalence of ATTR-CM

ATTR-CM is a rare disease with heterogeneous epidemiology across geographic regions and populations. Depending on the population studied and the diagnostic method used, ATTR-CM prevalence ranges from 6.1 per million in the US to 50 per million in Sweden, and Japan (100 per million per year) [6] (Figure 3).

Figure 3: Worldwide prevalence of ATTR amyloidosis.



Except for the data from Japan and Portugal, all other prevalence and incidence rates depicted in this figure pertain to unspecified ATTR-CM. Abbreviations: ATTR-PN=amyloid transthyretin polyneuropathy; PMPY=Per Million Per Year; ATTRwt-CM=wild-type amyloid transthyretin cardiomyopathy

Source: Figure adopted from Delgado, et al. 2025 [6].

Since the introduction of nuclear cardiac imaging with technetium pyrophosphate scan, the diagnosis of ATTR-CM is possible without cardiac biopsy facilitating an earlier diagnosis and treatment resulting in improved survival rates and higher prevalence numbers. In general, a substantial higher prevalence and incidence of ATTR-CM is observed among men, the elderly (≥ 65-years), and in people with cardiomyopathy and/or progressive heart failure. In a pooled analysis from 69 studies and 4669 patients with ATTR-CM, 17% were females, and 83% were men. Studies of ATTRwt had the lowest proportion of females (9%), whereas ATTRv had the highest (29%) [7].

3.3. Amyloidosis - orphan drug designation

Drugs intended as therapy for a rare disease may receive a so-called 'orphan drug designation' by regulatory bodies to incentivize their development. In 1983, the Orphan Drug Act (ODA) defined a rare disease as a disease or condition that affects less than 200,000 people in the United States. In Europe, Regulation (EC) No 141/2000 established criteria for designating a medicinal product as an orphan medicinal product (OMP) within the European Union. EMA is responsible for reviewing applications from sponsors for orphan designation.

To qualify for orphan designation, a medicine must meet a number of criteria:

- **Seriousness:** It must be intended for the treatment, prevention or diagnosis of a disease that is life-threatening or chronically debilitating;
- **Rarity:** The prevalence of the condition in the EU must not be more than 5 in 10,000 or it must be unlikely that marketing of the medicine would generate sufficient returns to justify the investment needed for its development;
- **Alternative methods:** No satisfactory method of diagnosis, prevention or treatment of the condition concerned can be authorized, or, if such a method exists, the medicine must be of significant benefit to those affected by the condition.

In 2011, an 'orphan designation' (based on designation EU/3/06/401) was assigned to tafamidis for the treatment of familial amyloid polyneuropathy. The 10 years orphan market exclusivity for this indication ended in 2021.

A 10-years orphan market exclusivity for treatment of senile systemic amyloidosis (based on designation EU/3/12/1066) started on 19 Feb 2020 [8]. In 2012, orphan designation of tafamidis had also been granted in the U.S.A for symptomatic transthyretin amyloid cardiomyopathy (AATR-CM) that is still active.

In 2019, tafamidis meglumine (Vyndaqel) and tafamidis (Vyndamax) received FDA marketing authorization for the treatment of ATTR-CM for both the wildtype (ATTRwt) and mutant variants (ATTRv) of ATTR amyloidosis. Shortly thereafter, tafamidis received a centralized EU marketing authorisation under the name Vyndaqel on 19 Feb 2020 (with the number EU/1/11/717) for the treatment of ATTRwt and ATTRv amyloidosis in adult patients with cardiomyopathy (ATTR-CM). On country levels the orphan drug designation depends on health care budgets and might judge the impact differently.

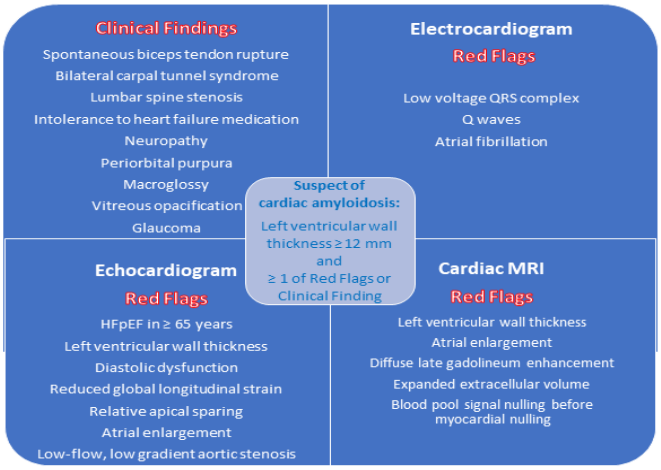
Unfortunately, the unfavorable cost analysis of tafamidis with high annual treatment cost of US$225,000 is limiting its access in many countries [9].

3.4. Management of cardiac amyloidosis

The symptoms of ATTR-CM can mimic various conditions such as other types of cardiomyopathies and heart failure. If ATTR-CM remains untreated, heart failure may worsen rapidly and associated dysfunctions of the cardiac conduction system may cause sudden cardiac death due to fatal arrhythmias or complete heart block. ATTR-CM is still heavily underdiagnosed because of lack of knowledge or lack of effective cardiac imaging techniques. The disease is also often misdiagnosed because it may mimic hypertrophic cardiomyopathy or ischemic heart disease. It is also often unrecognized in older individuals with severe aortic stenosis undergoing trans catheter aortic valve replacement. In addition, there are no biomarkers to estimate future cardiac involvement in individuals who are initially asymptomatic. A further difficulty for diagnosis is that the clinical manifestations of ATTR neuropathy (ATTR-PN) or ATTR-cardiomyopathy (ATTR-CM) often overlap depending on the mutation variant, age, gender and other factors [5].

Systemic amyloidosis can affect many organs and organ systems and can be associated with visual, neurological, dermatological, cardiac, renal and orthopedic signs and symptoms. Several clinical and non-invasive diagnostic assessments (electrocardiogram, echocardiography, cardiac MRI) can be used to identify 'Red Flags' that suggest a patient may have cardiac amyloidosis (Figure 4). Increased left ventricular wall thickness in an undilated left ventricle is a feature of cardiac amyloidosis and should prompt further investigation in elderly patients with heart failure with preserved ejection fraction, hypertrophic cardiomyopathy or severe aortic stenosis, particularly those undergoing trans catheter aortic valve replacement. In 2015, striking findings in elderly patients (> 60 years) with HFpEF (left ventricular ejection fraction of more than 50% with LV hypertrophy of more than 12 mm) were reported demonstrating that ATTR-wt accounts for a significant number (13%) of HFpEF cases [10]. In the case of ATTR-wt, the normal (non-mutated) transthyretin tetramer protein easily dissociates into misfolding monomers, which over a period of decades gradually deposit as amyloid fibrils in cardiomyocytes. In this study, all elderly HFpEF patients with a positive (99m) Tc-DPD scintigraphy underwent genetic testing. When no transthyretin mutations were found, ATTR-wt could be confirmed by myocardial biopsy. Similar findings were reported later by other cardiologists [11,12]. The remarkably high number of HFpEF patients with the ATTR-wt genotype (i.e., phenotype ATTR-CM) in these studies led to the recommendation that this entity should be routinely considered in all HFpEF patients.

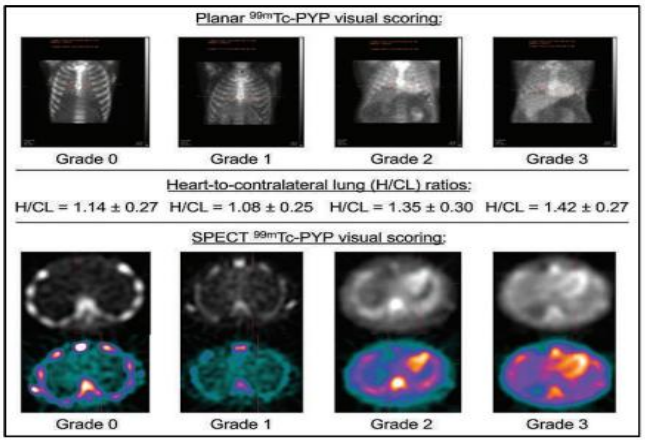
Figure 4: Clinical findings and red flags suspecting cardiac amyloidosis.



As light-chain amyloidosis (AL) is a rapidly progressive and fatal disease requiring timely chemotherapy, an important step in diagnosing cardiac amyloidosis is to rule out AL disease. Monoclonal proteins indicative for AL can be detected by a serum and urine immunofixation electrophoresis and serum free light-chain assay (SPIE, UPIE and SFLC, respectively). Laboratory testing for monoclonal proteins is performed in conjunction with ^{99m}Tc-pyrophosphate (^{99m}Tc-PYP) imaging to assess cardiac amyloid deposition.

Cardiac amyloidosis can be quantified by planar and SPECT ^{99m}Tc-PYP visual scoring [13] (Figure 5). The mechanism of ^{99m}Tc-PYP binding to amyloid deposits is probably related to the calcium-dependent P-component of amyloid fibrils, which facilitates the uptake and binding of bone-seeking radiopharmaceuticals to amyloid deposits, allowing visualization [14].

Figure 5: Cardiac amyloidosis ^{99m}Tc-PYP semi quantitative and quantitative interpretation.



(Top) Planar 3-h images demonstrating diffuse myocardial uptake and grade based on comparison of 99mTc-pyrophosphate myocardial uptake with rib. (Middle) H/CL ratio. (Bottom) Gray-scale and color SPECT images confirming myocardial uptake. Abbreviation: PYP = pyrophosphate.

Source: Figure adopted from Jerome, et al. 2023 [13]

Based on the outcomes of these diagnostic tests, the following differential diagnoses are possible:

When monoclonal proteins are present

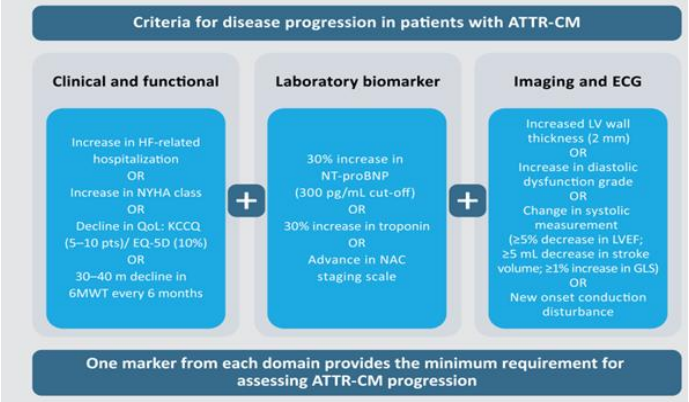
- and ^{99m}Tc-PYPnuclear imaging is positive (grade 2or 3), cardiac AL is likely and the patient should undergo further haemato-oncologic evaluation. If monoclonal proteins are present and ^{99m}Tc-PYP results are ambiguous (grade 1), additional histologic assessment is required for diagnosis.
- and ^{99m}Tc-PYP scan is negative (no myocardial tracer uptake), AL-CM can be excluded, but further assessments are needed to confirm systemic AL.

When monoclonal proteins are absent

- and ^{99m}Tc-pyrophosphate scan demonstrates myocardial uptake, findings indicate cardiac ATTR, and the patient should undergo genetic testing to determine type of cardiac amyloidosis (ATTR-wt or ATTRv).
- and there is no ^{99m}Tc-PYP uptake in the myocardium, cardiac AL and cardiac ATTR are unlikely.

In 2021, experts of the *European Society of Cardiology* agreed about a consensus document recommending a set of clinically feasible tools for the diagnostics and long-term monitoring of patients with ATTR-CM, including meaningful thresholds for defining disease progression and the frequency of measurements [15] (Figure 6).

Figure 6: Expert consensus on the long-term monitoring of patients with ATTR-CM.



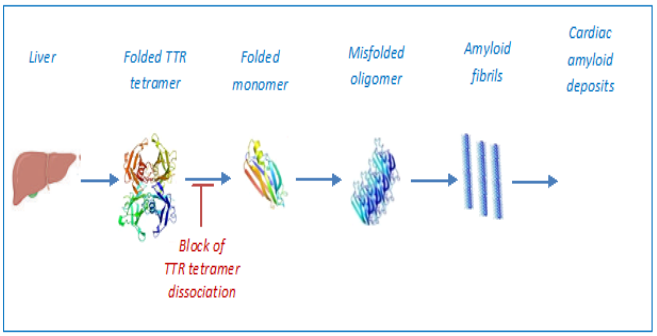
Abbreviations: ATTR-CM=transthyretin amyloid cardiomyopathy, 6MWT = 6-min walk test, ECG = electrocardiogram, EQ-5D = EuroQol five dimensions, GLS = global longitudinal strain, HF = heart failure, KCCQ = Kansas City Cardiomyopathy Questionnaire, LV = left ventricular, LVEF = left ventricular ejection fraction, NAC = UK National Amyloidosis Centre, NT-pro BNP = N-terminal pro-B-type natriuretic peptide, NYHA = New York Heart Association, QoL = quality of life.

Source: Figure adopted from Garcia-Pavia, et al. 2021 [15].

4. Approved Disease Modifying TTR-specific Therapies

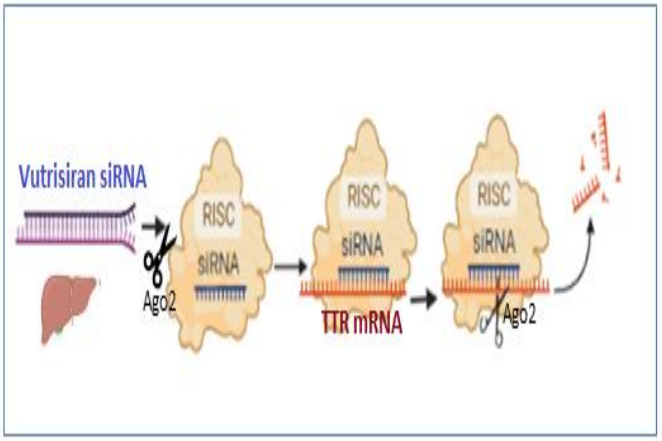
Once ATTR-CM diagnosis is confirmed, it requires a highly individualized treatment including management of cardiac symptoms (due to heart failure or arrhythmias) and treatment of the underlying amyloidosis. In case of heredity TTR amyloidosis (ATTRv), a liver transplant can remove mutant TTR from circulation. Since the advent of TTR-specific therapies, the need for liver transplants has however dramatically reduced. Emerging therapeutic strategies for ATTR-CM are targeting the mechanisms of amyloid deposition in the myocardium. Disease-modifying TTR-specific therapies have been approved that either stabilize the TTR homotetramer to prevent tissue deposition (tafamidis and acoramidis) (Figure 7) or block the synthesis of mutated TTR protein by mRNA silencing (vutrisiran) (Figure 8).

Figure 7: TTR stabilizer tafamidis and acoramidis reduce TTR tetramer dissociation.



TTR homotetramer, misfolding monomers, fibril formation, and deposition are depicted for ATTR-CM. In both wildtype (ATTRwt) and hereditary (ATTRv) ATTR-CM, transthyretin tetramers are secreted by liver, fold abnormally, and form amyloid fibrils that are deposited in the heart.
Created by BioRender.com

Figure 8: Suppression of TTR mRNA translation in hepatocytes by vutrisiran.



Vutrisiran siRNA binds to enzyme complex RISC (RNA-induced silencing complex), where it is unwound: the RISC protein AGO2 carries out cleavage of the sense strand allowing the antisense strand to bind its target TTR mRNA. Once the target RNA is bound to the antisense strand, its phosphodiester backbone is cleaved by AGO2. This leads to sequence-specific knockdown of TTR mRNA and causes TTR gene silencing. Abbreviations: AGO2 = argonaute 2, siRNA = small interfering RNA, TTR

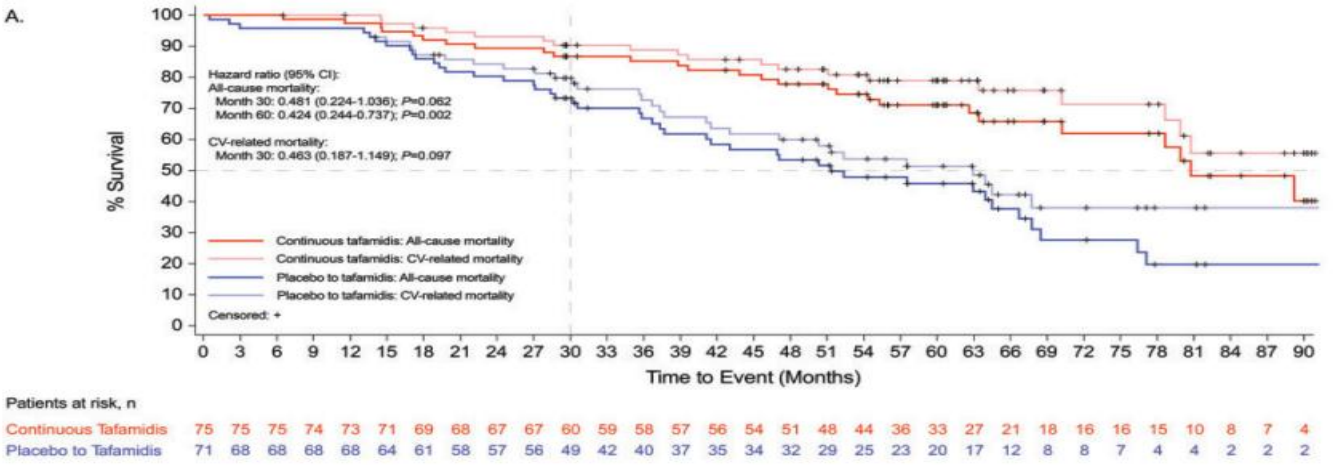
= transthyretin, RISC = RNA-induced silencing complex
Created by BioRender.com

4.1. TTR stabilizers

Tafamidis is derived from the non-steroidal anti-inflammatory drug (NSAID) diflunisal retaining selectively TTR-stabilizing properties but without cyclooxygenase inhibition and without unwanted NSAID side effects. Tafamidis is a selective stabilizer of TTR. It binds to TTR at the thyroxine binding sites, stabilizing the tetramer and slowing dissociation into monomers, the rate-limiting step in the amyloidogenic process.

Tafamidis was the first selective TTR stabilizer receiving marketing authorization by FDA (VYNDAMAX™, 2019) and EMA (VYNDAQEL®, 2020) for patients with hereditary or wildtype ATTR-CM. The approvals were given due to the results of the ATTR-ACT trial(NCT01994889)that enrolled 441 patients with heart failure (NYHA I-III) and histological confirmed ATTR-CM, randomized to 80 mg of tafamidis meglumine, 20 mg of tafamidis meglumine, or placebo in a 2:1:2 fashion and followed over 30 months. Treatment with tafamidis led on average to a 30% reduction in all-cause mortality compared to placebo (number needed to treat (NNT) of 7.5 to prevent one death). In addition, tafamidis treatment was associated with 32% reduction of cardiovascular-related hospital admissions (NNT of 4 to prevent one cardiovascular hospitalization per year). Approximately 80% of total deaths were cardiovascular-related in both treatment groups [16]. After completing ATTR-ACT, patients could receive tafamidis 61 mg up to 60 months in the open-label long-term extension (LTE) study (NCT02791230). Heart transplant and cardiac mechanical assist device implantation were treated as death in the mortality assessments. Patients were stratified according to the National Amyloidosis Centre (NAC) staging for ATTR-CM into prognostic categories. Continuous tafamidis treatment reduced mortality and hospitalizations (both all-cause and CV-related) across all NAC stages. These reductions were larger and occurred earlier in patients with NAC stage I, emphasizing the importance of early disease-modifying treatment in patients with ATTR-CM [17]. A Kaplan-Meier plot of time to event all-cause mortality and hospitalizations is presented in Figure 9.

Figure 9: All-cause and CV-related (A) mortality in NAC stage I ATTR-CM at baseline and (B) hospitalizations summary across all NAC stages in the ATTR-ACT (NCT01994889) and the LTE (NCT02791230) trials.



B.

	NAC Stage I		NAC Stage II		NAC Stage III	
	Placebo to Tafamidis n=71	Continuous Tafamidis n=75	Placebo to Tafamidis n=72	Continuous Tafamidis n=62	Placebo to Tafamidis n=34	Continuous Tafamidis n=36
Patients with all-cause hospitalisations, n (%)	31 (43.7)	44 (58.7)	21 (29.2)	27 (43.5)	7 (19.4)	9 (25.0)
Total all-cause hospitalisations per year[a]	0.70	0.46	0.60	0.33	0.79	0.57
Patients with CV-related hospitalisations, n (%)	23 (32.4)	29 (38.7)	15 (20.8)	19 (30.6)	5 (13.9)	6 (16.7)
Total CV-related hospitalisations per year[a]	0.62	0.36	0.49	0.27	0.45	0.23

[a] Total all-cause or CV-related hospitalisations across all participants divided by total years of study participation across all participants.
CV=cardiovascular; NAC=National Amyloidosis Centre

Source: Figure adopted from Damy, et al. 2024 [17].

Analyses of real-world post-marketing (FDA pharmacovigilance) data for tafamidis identified potential side effects including hearing system damage and urinary tract infections, but no signals of potentially fatal or disabling adverse reactions confirming a favorable safety profile for tafamidis [18].

In the ATTR-ACT trial, TTR kinetic stability reached maximum with higher tafamidis plasma concentrations pointing to an optimum dose of 80 mg tafamidis meglumine (4 capsules). The subsequent development of a single capsule containing tafamidis 61 mg free acid (bioequivalent to tafamidis meglumine 80 mg) simplified the number of pills to be taken.

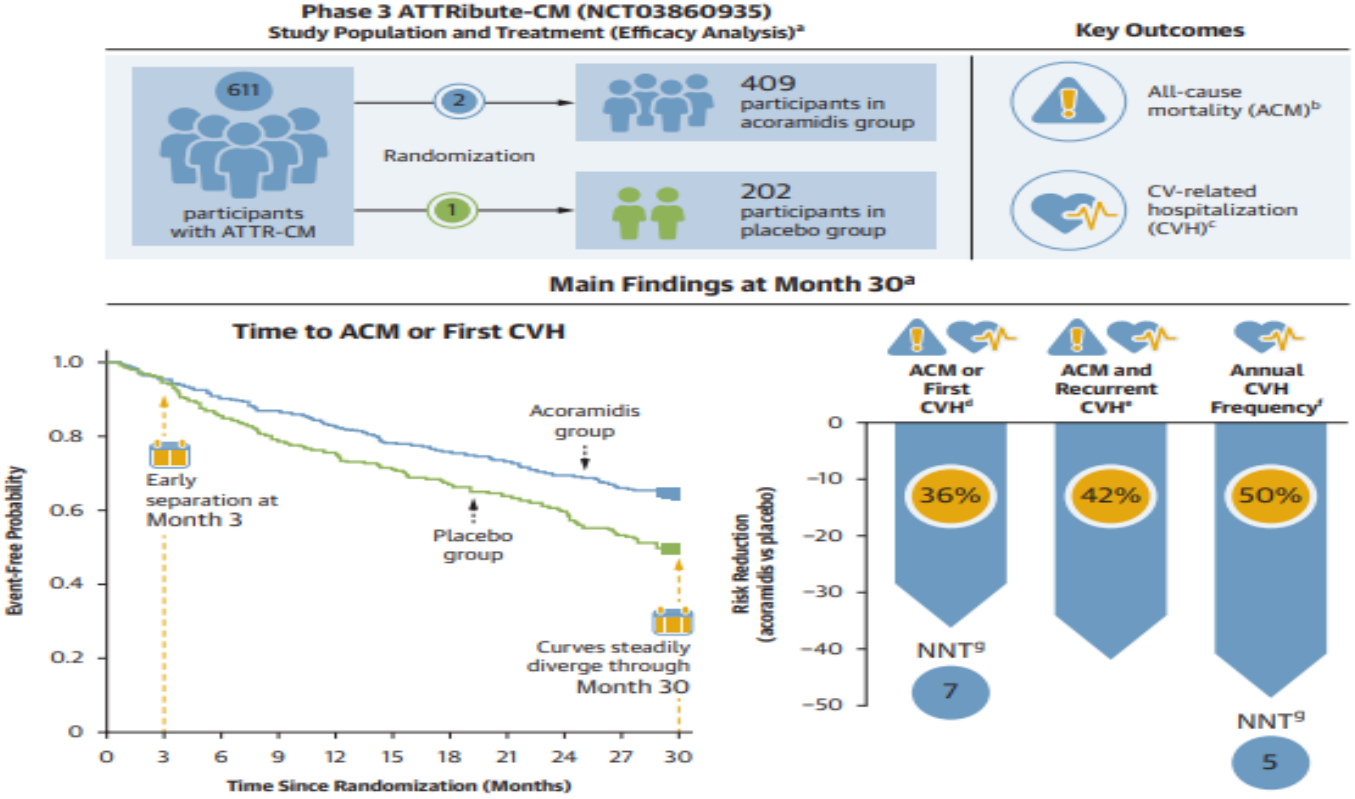
The current ESC CHF guidelines include a class I recommendation for tafamidis for the treatment of hereditary and wildtype ATTR-CM and NYHA class I or II to reduce symptoms, CV hospitalizations, and mortality as a class I recommendation [19]. The 2022 ACC/AHA/HFSA guidelines also recommend tafamidis for selected patients with NYHA class III. Benefit has not been observed in heart failure patients with NYHA IV symptoms, severe aortic stenosis, or impaired renal function (eGFR < 25 $mL \cdot min^{-1}$ 1.73 m^{-2} body surface area) [20,21].

Acoramidis (formerly called AG10) is a next generation TTR stabilizer and represents an important expansion of treatment options for patients with cardiac amyloidosis.

Acoramidis mimics the natural protective T119M mutation, which shows a 37-fold greater stabilization of TTR than in wild type [22]. *In vitro* and *ex vivo* assessments of acoramidis have shown TTR stabilization that is near-complete (~90% across the entire dosing interval) and greater than that achieved with therapeutic concentrations of tafamidis [23,24].

In a phase 2 trial, AG10 400 mg or 800 mg taken twice daily for 28 days normalized serum TTR levels in all ATTR-CM patients with symptomatic, chronic heart failure regardless of baseline and mutation status [24]. Acoramidis received marketing authorisation by FDA (ATTRUBY™, 2024) and EMA (ATTRUBY®, 2025) due to the results of the ATTRibute-CM trial (NCT03860935) including patients with NYHA Class I-III symptoms due to ATTR cardiomyopathy. Acoramidis showed clinical efficacy compared with placebo for the primary outcome: a hierarchical 4-component analysis of all-cause mortality (ACM), the cumulative frequency of cardiovascular hospitalization (CVH), the change from baseline in N-terminal pro–B-type natriuretic peptide (NT-proBNP) level, and the change from baseline in 6-minute walk distance (6MWD) over 30 months. Acoramidis also showed a superior treatment effect compared with placebo for the prespecified secondary endpoint of the 2-component hierarchy of ACM and CVH [25,26] (Figure 10). Preliminary data from the ATTRibute-CM open-label extension (OLE) trial for acoramidis show a relative risk reduction in all-cause mortality of 34% vs. placebo after 42 months [27].

Figure 10: Benefits of acoramidis on all-cause mortality and cardiovascular-related hospitalization in ATTR-CM.



[a]Efficacy analyses were conducted in the modified intention-to-treat population (participants with one postbaseline efficacy evaluation and a baseline estimated glomerular filtration rate ≥ 30 mL/min/1.73 m^2).
[b]Death from any cause, heart transplantation, or implantation of a cardiac mechanical assist device.
[c]Nonelective admission to an acute care setting for cardiovascular (CV)-related morbidity that resulted in a ≥ 24-hour stay, or an unscheduled medical visit of < 24 hours owing to heart failure and requiring treatment with intravenous diuretics.
[d]Risk reduction is calculated as 1 - HR of the Kaplan-Meier analysis and is expressed as a percentage.
[e]Analyzed using negative binomial regression.
[f]Risk reduction is calculated as 1 - relative risk ratio of the annual cardiovascular-related hospitalization (CVH) frequency occurring in participants treated with acoramidis vs placebo and is expressed as a percentage.
[g]Number-needed-to-treat (NNT) is the number of participants required to be treated for 30 months for one participant to experience a treatment benefit compared with placebo. ACM = all-cause mortality; ATTR-CM = transthyretin amyloidosis cardiomyopathy.

Source: Figure adopted from Judge, et al. 2025 [26].

4.2. TTR gene silencer

Vutrisiran is a chemically modified double-stranded small interfering ribonucleic acid (siRNA) that targets mutant and wild-type transthyretin (TTR) messenger RNA (mRNA) and is covalently linked to a ligand containing three N-acetyl galactosamine (GalNAc) residues to enable delivery of the siRNA to hepatocytes. The phosphorothioate backbone utilized in vutrisiran (instead of the phosphodiester backbone used in patisiran) enhances hepatocyte delivery and uptake, stabilizes the molecule during systemic circulation and is associated with a better tolerability. The improved stabilization chemistry of vutrisiran and other modifications (increased 2′-O-methyl nucleotide content) enables subcutaneous administration once every 3 months [28,29].

Treatment with 25 mg vutrisiran administered by subcutaneous injection every 3 months results in a rapid and significant reduction of serum TTR protein as has been shown in the two phase 3 HELIOS-A (NCT03759379) and HELIOS-B (NCT04153149) trials. In HELIOS-A (including adult patients with heredity ATTR-PN), vutrisiran reduced mean serum TTR at steady state by 83% and 88% after 9 and 18 months of treatment, respectively [29]. Similar TTR

reductions were observed regardless of Val30Met genotype status, weight, sex, age, or race. In HELIOS-B (including adult patients with hereditary ATTR-CM) the mean trough percent reduction was 81.0% (95% CI, 79.0 to 83.0) at 30 months in the overall population. In addition, treatment with vutrisiran led to a lower risk of death from any cause and cardiovascular events than placebo and preserved functional capacity and quality of life. Biomarkers associated with heart failure (NT-proBNP and Troponin I) favored vutrisiran over placebo. The incidence of adverse events was similar in the two groups [30]. Based on the results of HELIOS-B trial, vutrisiran received marketing authorisation by FDA (AMVUTTRA™, March 2025) and EMA (AMVUTTRA®, June 2025) as treatment for cardiomyopathy of wild-type or hereditary transthyretin-mediated amyloidosis (ATTR-CM) in adults to reduce cardiovascular mortality, cardiovascular hospitalizations and urgent heart failure visits.

Since vutrisiran treatment leads to a decrease in serum vitamin A levels (transthyretin is a vitamin A [retinol] carrier), patients need to take the recommended daily allowance of vitamin A. In case patients develop symptoms of vitamin A deficiency such as night blindness, they should be referred to an ophthalmologist.

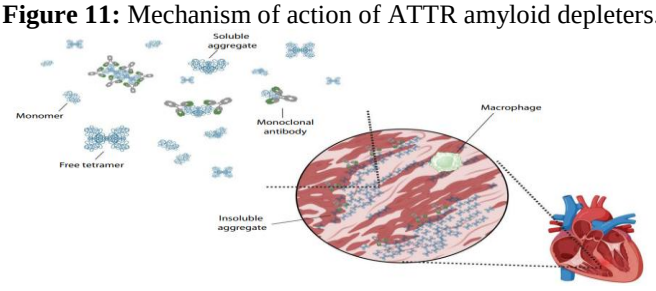
The annual costs of AMVUTTRA® therapy is as high as US$476,000 per year. That's more than double the cost of acoramidis (BEYONTTRA®) or tafamidis (VYNDAQEL®) for the same condition. Alnylam offers a patient support programme called 'Alnylam Assist' which may support patients if the insurance does not provide sufficient cover.

The current 'Disease Modifying Therapies' for ATTR-CM approved and available in the EU and the US are summarized in Appendix 2.

5. Drug Pipeline for ATTR-CM

Novel therapeutic strategies for the treatment of ATTR-CM are emerging addressing various aspects of the disease process. The novel strategies seek to inhibit TTR synthesis using RNA silencing methods, including the antisense oligonucleotide (ASO) eplontersen and the siRNA nucresiran or gene-editing agents using CRISPR/Cas9 technologies such as NTLA-2001 and YOLT-201. The promising gene editing approaches hypothesize that target DNA can be permanently modified requiring only a single (once-in-a-lifetime) drug dose. Other novel approaches are promoting the degradation and removal of toxic amyloid to restore cardiac physiology by using monoclonal antibodies such as ALXN2220, AT-02 and coramitug (called ATTR amyloid depleters).These monoclonal antibodies disrupt amyloid fibrils in cardiac tissue, aiding their elimination by phagocytic cells [31] (Figure 11). Patients experiencing symptomatic amyloid organ deposition that may not respond to TTR silencers or stabilizers may in particular benefit from this novel approach.

Figure 11: Mechanism of action of ATTR amyloid depleters.



Source: Figure adopted from Keller, et al. 2025 [31].

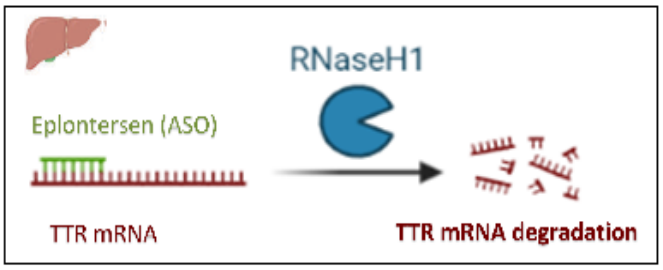
In the following, the current drug pipeline for ATTR-CM will be presented and discussed according to the development phase of each drug candidate.

5.1. Phase 3 drug candidates

5.1.1. Ongoing phase 3 projects:

Eplontersen (Ionis Pharmaceuticals/Astra Zeneca) is a longer acting antisense oligonucleotide (ASO) conjugated to GalNAc, which facilitates its uptake into hepatocytes via the asialoglycoprotein receptor and increases the efficacy of the drug. Eplontersen causes degradation of mutant and wild-type TTR mRNA through binding to the TTR mRNA and by ribonuclease H1 (RNase H1), which results in a reduction of serum TTR protein and TTR protein deposits in tissues (Figure 12).

Figure 12: Suppression of TTR mRNA translation in hepatocytes by eplontersen.



Eplontersen is an RNase H1 dependent ASO which binds complementary to TTR mRNA thereby inducing post-transcriptional gene silencing. RNase H1 is an endogenous enzyme that recognizes DNA–RNA heteroduplexes bound to their complementary target mRNA strand and catalyzes the degradation of the targeted mRNA.
Created by BioRender.com

Eplontersen has already been approved by FDA (2023) as WAINUA™ and by EMA (2024) as WAINZUA® for the treatment of ATTR-polyneuropathy (ATTR-PN) in adults with mutant variants of ATTR amyloidosis (ATTRv). Eplontersen treatment leads to a decrease in serum vitamin A levels (transthyretin is a vitamin A [retinol] carrier). Before initiating treatment with eplontersen, serum vitamin A levels below the lower limit of normal must be corrected and any ocular symptoms or signs suggestive of vitamin A deficiency must be investigated. In 2024, eplontersen received an FDA Fast Track Designation for the treatment of ATTR-CM.

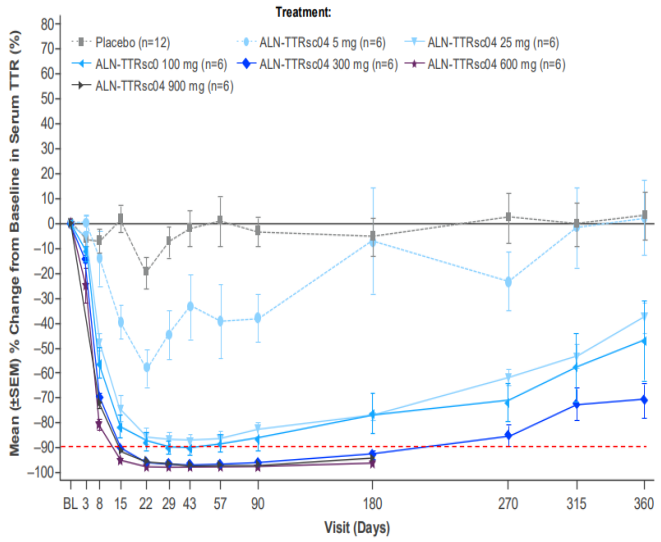
Eplontersen is currently investigated in the phase 3 CARDIO-TTRansform trial (NCT04136171)to evaluate the efficacy of eplontersen compared to placebo in participants with ATTR-CM receiving available standard of care therapy. The global CARDIO-TTRansform trial is already fully enrolled with more than 1,400 patients. Study completion is expected in Q2 2026.

Nucresiran (formerly ALN-TTRsc04) from Alnylam Pharmaceuticals is a third generation siRNA using IKARIATM, an advanced platform methodology, to identify sequences with improved potency and durability as well as target specificity. Nucresiran blocks the synthesis of both wildtype and mutated TTR protein by mRNA silencing.

In a Phase 1 study (NCT05661916), a single 300 mg subcutaneous dose of nucresiran led to TTR knockdown > 90% by Day 15 and peak knockdown > 96% by Day 29, which was maintained for over 6 months with low inter subject variability [32] (Figure 13). Nucresiran was safe and well tolerated; the majority of adverse events were mild in

severity and none were considered treatment-related.

Figure 13: Mean percent change from baseline in serum TTR levels over time.



Abbreviations: ALN-TTRsc04 = nucresiran, BL= baseline, SEM = standard error of the mean, TTR = transthyretin

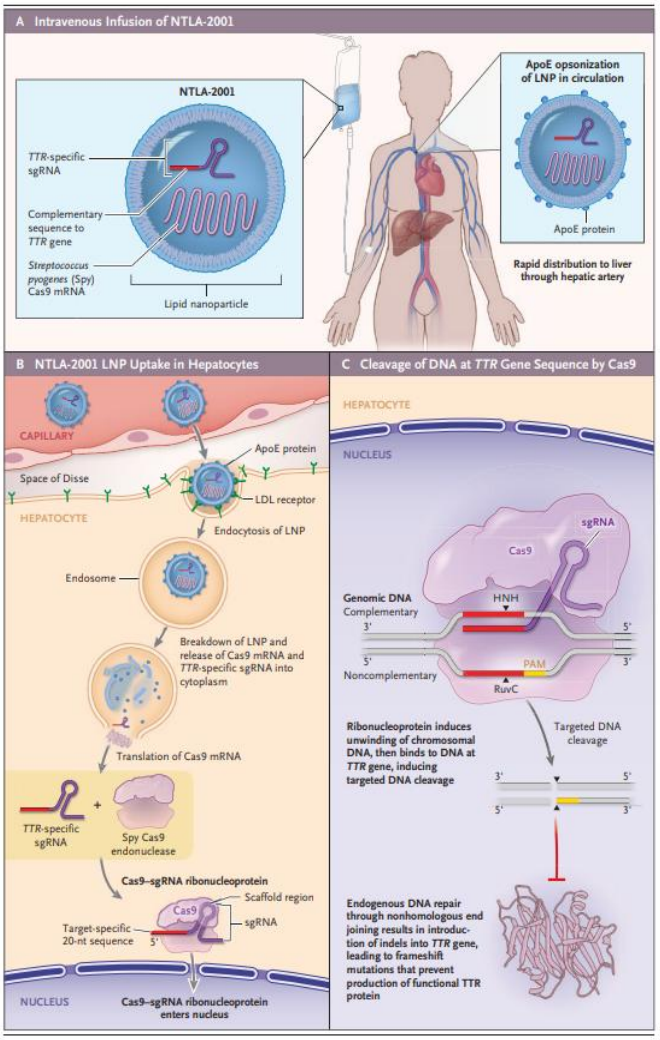
Source: Figure adopted from Murad, et al. 2024 [32].

In July 2025, a global, randomized, double-blind, event-driven phase 3 trial (NCT07052903, TRITON-CM) commenced that will investigate the efficacy and safety of nucresiran in 1250 patients with ATTR-CMof NYHA class I-III. This trial tests the hypothesis that nucresiran can reduce all-cause mortality and recurrent cardiovascular events in patients with ATTR-CM. Participants will receive nucresiran 300 mg or placebo administered subcutaneously (SC) once every 6 months (q6M) during the 24 months double-blind period, and the following open-label extension period. Heart failure patients with poor prognosis such as NYHA class IV, NYHA class III with ATTR Amyloidosis Disease Stage 3 or Polyneuropathy Disability Score ≥ IIIa, and patients with estimated glomerular filtration rate (eGFR) < 30mL/min/1.73 m2) will be excluded. Primary study completion is expected in 2030.

ALXN2220 (formerly NI006) from Alexion Pharmaceuticals is a recombinant human anti-ATTR monoclonal antibody designed to deplete cardiac ATTR amyloid deposits in patients with ATTR-CM on top of standard of care. In the *first-in-human* trial NI006-101, the use of NI006 was not associated with any apparent drug-related serious adverse events, and its pharmacokinetic profile resembled that of an IgG antibody, with no antidrug antibodies detected. Over 12 months, cardiac tracer uptake on scintigraphy and extracellular volume on cardiac magnetic resonance imaging, both imaging-based surrogate markers of cardiac amyloid load, appeared to decrease. In a prolonged Open-Label Extension (OLE) of the NI006-101 study, long-term treatment with ALXN2220 at 30 mg/kg was safe, well tolerated, and showed high adherence, with no severe adverse events, no events of cytokine release syndrome or thrombocytopenia, and no anti-drug antibodies were detected. Cardiac imaging and biomarker data demonstrated sustained treatment effects beyond 12 months, supporting dose up-titration to 30 mg/kg, with participants experiencing continued reductions in cardiac biomarkers and imaging supported efficacy over a median follow-up of 125 weeks [33]. A global phase 3 multicenter study called DepleTTR-CM (NCT06183931) is currently conducted to assess the safety and efficacy of ALXN2220 in adult patients with ATTR-CM. The primary outcome measure focuses on the difference between ALXN2220 and placebofor total occurrences of all-cause mortality and cardiovascular clinical events during the blinded treatment period (up to 48 months).

Nexiguran-ziclumeran (nex-z, formerly NTLA-2001) from Intellia Therapeutics is an *in vivo* gene-editing therapeutic agent designed to inactivate the *TTR* gene that encodes for the mutated TTR protein. This novel CRISPR (clustered regularly interspaced short palindromic repeats) and combined Cas9 (Cas9 endonuclease) system comprises a lipid nanoparticle encapsulating mRNA for Cas9 protein and a single guide RNA (sgRNA) targeting the TTR gene [34] (Figure 14).

Figure 14: Mechanism of action of Nexiguran-ziclumeran (nex-z, formerly NTLA-2001).



After single intravenous infusion, the two-component genome editing system encapsulated by lipid nanoparticles is opsonized by apolipoprotein E and transported directly to the liver, where it is taken up by LDL receptors on hepatocytes, followed by endocytosis of the particle. Upon gradually release into the cytoplasm, the Cas9 mRNA molecule is ribosomal translated resulting in the Cas9 endonuclease enzyme that interacts with the TTR-specific sgRNA, forming a CRISPR–Cas9 ribonucleoprotein complex that enters the nucleus. This complex enters the nucleus, where the sgRNA binds to the complementary sequence of the TTR DNA sequence. Endogenous DNA repair subsequently leads to indels (insertions or deletions of bases) in the TTR gene that prevent the production of the TTR protein (knockout mutations).

Source: Figure adopted from Gill more, et al. 2021 [34].

Preliminary phase 1 data are showing that a single dose of nex-z led to consistently rapid, deep and durable reduction in serum TTR [35]. Nex-z has been granted Regenerative Medicine Advanced Therapy designations by the FDA for both cardiomyopathy and polyneuropathy. Nex-z has also been granted Orphan Drug Designation by the FDA and European Commission.

The global pivotal phase 3 MAGNITUDE trial (NCT06128629) seeks to enrole 765 patients with ATTR-CM to evaluate the efficacy and safety of a single infusion of NTLA-2001 or placebo as a one-time treatment. The primary endpoint is a composite outcome of cardiovascular mortality and cardiovascular events over 18 months and (depending on event rates) up to 48 months.

5.1.2. Recently declined phase 3 project: Patisiran from Alnylam was the first FDA-approved (2019) RNA silencing therapy for hereditary ATTR with polyneuropathy. Itis a double-stranded siRNA (formulated as lipid nanoparticles for delivery to hepatocytes) targeting a genetically conserved sequence in the 3'-untranslated region (3'-UTR) of both mutant and wild-type TTR mRNA, leading to direct, sequence-specific degradation of TTR mRNA in the liver [36].

In the phase 3 APOLLO-B trial (NCT03997383), administration of patisiran over a period of 12 months exhibited superiority over placebo in preserving functional capacity and improving quality of life endpoints in patients with ATTR-CM. Infusion-related reactions, arthralgia, and muscle spasms occurred more often among patients in the patisiran group than among those in the placebo group [37]. However, the FDA rejected a supplemental new drug application for patisiran to treat wild-type or hereditary ATTR-CM due to insufficient evidence of clinical meaningfulness. In 2023, Alnylam announced the discontinuation of efforts to seek approval for the extended indication of patisiran in the US. Nevertheless, they remain committed to ensuring continued access to the product for ATTR-CM patients participating in the open-label extension (OLE) phase of APOLLO-B and through the expanded access protocol. Study completion is expected in March 2027.

5.2. Phase 2 drug candidates

AT-02 from Attralus Inc., is a humanized IgG1 monoclonal antibody genetically fused to the pan-amyloid binding peptide p5R. AT-02 demonstrates high binding potency to all types of amyloid deposits (ATTR and light chain amyloid fibrils), facilitating macrophage-mediated phagocytosis, enhanced by complement [38]. In a mouse model of systemic amyloidosis, AT-02 removed amyloid deposits in key organs including a near 50% reduction in cardiac amyloid [39]. Phase 1 (NCT05521022) and phase 2 (NCT05951049) trials are currently investigating the safety and tolerability of AT-02 in healthy volunteers and patients with systemic amyloidosis including ATTR-CM. Treatment with ATTR stabilizers and gene silencers can be continued during study participation.

Coramitug (also known asNNC6019-0001 or PRX004) from Novo Nordisk is a humanized monoclonal antibody designed to deplete amyloid via antibody-mediated phagocytosis bytargeting a specific epitope exclusively exposed on misfolded monomeric and aggregated forms of TTR (ATTR amyloid depleter) [40].

In a global phase 1 trial (NCT03336580), 21 patients with hereditary amyloidosis (ATTRv) completed the dose-escalation phase receiving PRX004 up to 30 mg/kg intravenously every 4 weeks for 3 months. A total of 17 patients could receive up to 15 additional doses in the Long-Term Extension (LTE) study. PRX004 was well tolerated at all doses, with dose-proportional exposure. Global Longitudinal Strain (GLS) and Neuropathy Impairment Score (NIS) were improved or maintained in 7 patients who received doses of ≥ 3 mg/kg for at least 9 months supporting its further clinical development [41].

A phase 2 trial (NCT05442047) assessed its potential in 99 ATTR-CM patients who have been randomized to receive coramitug at doses of 10 and 60 mg/kg, or placebo. Patients received monthly infusions for 12 months.The primary outcomes are the changes in 6-minute walk test (6MWT) and N-terminal-pro brain natriuretic peptide (NTproBNP). This study has been completed in May 2025 and study results are expected in 2025. Study participants who have completed this study (regardless whether they received coramitug or placebo) are now recruited in the open-label extension (OLE) trial NCT06260709 that now assesses the long-term safety of coramitug given intravenously 4 weeks for up to 35 months. In both phase 2 trials, treatment with ATTR stabilizers and gene silencers was/is allowed during study participation.

Genistein is a soybean isoflavone with anti-inflammatory and antioxidant properties that was discovered using in silico modelling. The growing legalization of the illicit drug marijuana will probably increase its use and its side effect on the cardiovascular system. The psychedelic component of marijuana Δ9 -tetrahydrocannabinol (Δ9 -THC) causes also vascular inflammation, oxidative stress, and atherosclerosis via cannabinoid receptor 1 (CB1) whereas CB2 activation is anti-atherogenic [42,43]. Genisteinantagonizes CB1 in the peripheric vasculature without relevant CNS penetration and hence less psychiatric side effects. Genistein has been shown to block harmful cardiovascular effects of Δ9 -THC while preserving clinically useful effects such as sedation and analgesia [44]. Genistein is currently tested in the phase 1/2a GASPAR trial (NCT06634108) in patients with heart failure due to ATTR-CM as an adjunct therapy alongside the standard of care, tafamidis. By targeting both the inflammatory mechanisms implicated in heart failure and the molecular pathogenesis of ATTR-CM, the study positions genistein as a potential modifier of disease progression. The GASPAR trial is focused on its impact on inflammatory and cardiometabolic biomarkers, along with the effects on cardiac function (echocardiography) and exercise capacity (6-minute walk test). The study is sponsored by the London Health Sciences Centre Research Institute.

5.3. Phase 1 Drug Candidates

YOLT-201 from YolTech Therapeutics is a gene-editing drug utilizing an innovative lipid nanoparticle system to deliver CRISPR-Cas encoding mRNA and a guide RNA targeting the transthyretin-encoding TTR gene to human hepatic cells. By inactivating the TTR gene, YOLT-201 has the potential to reduce misfolded TTR protein production and lower serum TTR protein levels, addressing the underlying cause of ATTR-CM.

YOLT-201 is currently investigated in a single-arm, open-label, single-dose escalation investigator-initiated trial (NCT06082050) aimed at evaluating the safety, tolerability, pharmacodynamics and preliminary efficacy (6-minute walk test (6MWT), Kansas City Cardiomyopathy Questionnaire (KCCQ), cardiac biomarkers, echocardiography, cardiac MRI) of YOLT-201 treatment in patients with ATTR-CM.

Interim results from this trial were communicated in March 2025 reporting that patient enrolment was completed in 2024, with a total of 7 patients enrolled across 4 dose levels. In all dose levels, YOLT-201 was generally well-tolerated, with the most common treatment-related adverse events being infusion-related reactions (e.g. fever) and transient liver enzyme elevation. No adverse events led to treatment discontinuation. Patient enrolment was completed in 2024, with a total of 7 patients enrolled across 4 dose levels. In all dose levels, YOLT-201 was generally all well-tolerated, with the most common treatment-related adverse events being infusion-related reactions (e.g. fever), transient ALT/AST elevation. No adverse events led to treatment discontinuation. In the high-dose group, subjects achieved approximately 90% reduction in serum TTR after a single administration, with rapid, sustained, and stable declines, along with favorable safety and tolerability. For two patients in the low-dose group whose TTR reduction did not meet the target, a second administration was given, resulting in >95% reduction in serum TTR [45].

A tabular overview of the current global ATTR-CM drug pipeline available in the public domain at CLINICALTRIALS.GOV is given in Appendix 3.

5.4. Further potential drug candidates for ATTR-CM

CLR01 is a small molecule, originally developed as a biomimetic Lysine (Lys) receptor that binds Lys residues by a combination of hydrophobic and electrostatic interactions (lock-key model of molecular tweezers). The selective binding to Lys residues inhibits abnormal protein self-assembly preventing aggregation and decreasing the toxicity of multiple amyloidogenic proteins such as transthyretin [46]. LR01 has been tested in several *in vitro* and *in vivo* models of amyloidosis for inhibiting protein aggregation without signs of toxicity [47]. Further research is warranted to test whether CLR01 may prevent aggregation of transthyretin in ATTR-CM.

Diflunisal (generic) is a non-steroidal anti-inflammatory drug (NSAID) that confers enhanced stability to TTR tetramers as assessed by serum TTR levels that associate with attenuation of cardiac biomarker and left ventricular ejection fraction decline [48].

In a retrospective study at the Boston University Amyloidosis Center, diflunisal treatment for at least one year was associated with improved survival in patients with early stage wild-type ATTR-CM [49]. These promising study results need however to be confirmed in prospective, randomized clinical trials with larger patient cohorts. In any case, diflunisal might be a more cost-effective alternative to tafamidis or acoramidis for selected ATTR-CM patients with preserved renal function. At current, Purpose Pharma seeks a marketing authorisation for hereditary ATTR amyloidosis. In April 2025, the Committee for Medicinal Products for Human Use (CHMP) of the EMA has adopted a positive opinion for diflunisal (Attrogy® from Purpose Pharma) for the treatment of ATTRv in adult patients with stage 1 or stage 2 polyneuropathy. Diflunisal has been granted Orphan Drug Designation (ODD) for the treatment of ATTR amyloidosis in the EU. An application for ODD in the US has been submitted. Off-label use of diflunisal may be considered in wildtype ATTR-CM in combination with a proton pump inhibitor [19].

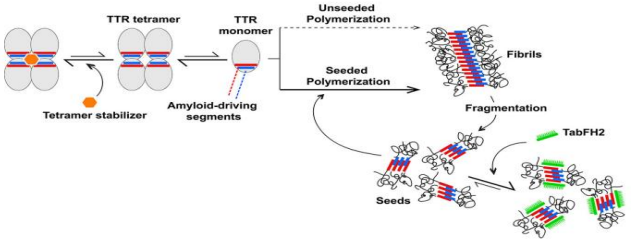
Doxycycline is a tetracycline antibiotic that has been shown to eliminate amyloid deposits when combined with the biliary acid tauroursodeoxycholic acid (TUDCA) or Ursodeoxycholic acid (UDCA) in transgenic TTR mouse models [50,51]. In a phase 2 clinical trial (NCT01171859) it could be demonstrated that treatment with Doxycycline/TUDCA in patients with ATTR amyloidosis was well tolerated and was able to prevent progression of cardiac dysfunction. Since then two larger prospective, randomized controlled trials (NCT01855360 and NCT03481972) were conducted to assess the safety and efficacy of combined Doxycycline/TUDCA in ATTR and ATTR-CM. Study results have not been published so far.

Epigallocatechin-3-gallate (a green tea component) is the most abundant catechin found in green tea, has some stabilization effect on TTR but may also promote amyloid degradation by converting existing fibrils into more soluble non-fibril conformers in vitro [52,53]. The efficacy and safety of epigallocatechin-3-gallate has been investigated in two phase 2 trials in patients with cardiac amyloid light-chain (AL) amyloidosis who completed chemotherapy (NCT01511263 and NCT02015312). Study results have not been published yet. Based on its mode of action, epigallocatechin-3-gallate may also improve cardiac transthyretin amyloidosis, but this remains to be elucidated.

TabFH2 is a transthyretin aggregation blocker (TAB). It is a mixture of two optimized peptide inhibitors (TabF2 and TabH2) targeting the F and H β-strands of TTR (called amyloid seeds) that are important segments driving aggregation, fibrillogenesis and spread of amyloid deposits.TabFH2 does not affect tetramer stability. Instead, TabFH2 binds to seeds in both wild-type and mutant TTR seeds, hindering self-recognition and seeding.

A combination of TTR stabilizers such as tafamidis (that reduces tetramer dissociation and unseeded polymerization) with the seeding inhibitor TabFH2may potentially act synergistic [54] (Figure 15). This combination could be of particular advantage for ATTR-CM patients with advanced TTR deposition. TABFH2 has been tested in preclinical Drosophila models for ATTR polyneuropathy showing improved motor parameters and reduced TTR deposition [55]. Research in ATTR-CM is warranted for this promising seeding inhibitor therapy.

Figure 15: Model of TTR amyloid seeding and its inhibition.



Abbreviations: TabFH2 = transthyretin aggregation blocker FH2, TTR =transthyretin

Source: Figure adopted from Saelices, et al. 2019 [54].

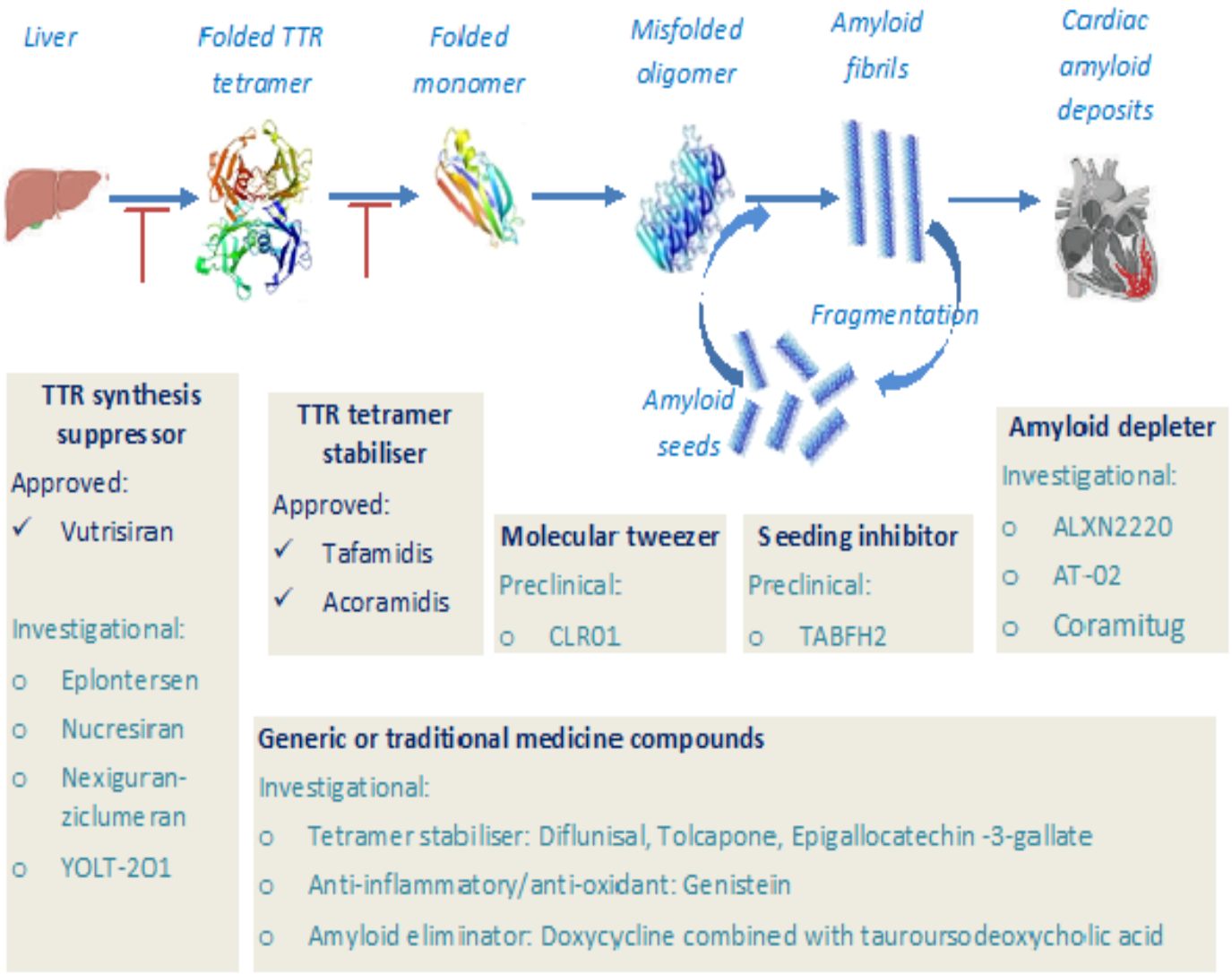
Tolcapone (formerly CRX-1008, SOM226) from Corino Therapeutics is a potent (small molecule) TTR stabilizer currently in phase 2 of development for the treatment of familial amyloid polyneuropathy (FAP). It is a repositioned compound that acts by imitating the process of binding thyroxine to TTR in the bloodstream and is administered via the oral route. It inhibits TTR tetramer dissociation into monomeric TTR and prevents the accumulation of amyloid in various tissues. Tolcapone has unique and differentiated activity compared to other TTR stabilizers, including the ability to cross an intact blood-brain barrier. Tolcapone may have potential to be also effective in ATTR-CM, but no clinical trials for this indication have been published so far.

6. Summary - Key Points

- Current approved TTR-specific therapies either block the formation of mutated TTR protein by mRNA silencing (vutrisiran) or stabilize the TTR tetramer to prevent amyloid tissue deposition (tafamidis, acoramidis) and have shown significant clinical improvements by reducing all-cause mortality, cardiovascular hospitalizations, heart failure biomarkers (NT-pro-BNP) and functional decline (6MWD) in patients with ATTR-CM. In many countries, access to these treatments is limited by their actual high annual treatment costs, which will hopefully decrease over time.
- Novel therapeutic approaches are emerging addressing various aspects of the ATTR-CM disease process such as inhibition of TTR synthesis either by RNA silencing using the eplontersen and nucresiran or by CRISP/Cas9 technology using the gene-editing agents nexiguran-ziclumeran or YOLT-201. Another novel approach promotes amyloid TTR degradation and removal, which includes the use of monoclonal antibodies such as ALXN2220, AT-02 or coramitug (so-called ATTR depleters).
- Novel strategies at preclinical stage include seeding inhibitors such as TabFH2 and molecular tweezers like CLR01 which inhibit TTR fibril formation.
- Generic and traditional medicine compounds, such as diflunisal, doxycycline combined with tauroursodeoxycholic acid, genistein, epigallocatechin-3-gallate (a green tea component) or tolcapone are further potential drug candidates for ATTR-CM.

Current approved therapies and novel emerging approaches are summarized in Figure 16.

Figure 16: Approved and emerging therapies for ATTR-CM**.**



Created by BioRender.com

7. Declarations

Ethics approval and consent to participate: Not applicable.
Consent for publication: All Authors have given their consent for publication.
Availability of data and materials: Not applicable.
Competing interests: All authors declare no competing interests.
Funding: Not applicable.

Authors' contributions:

- Conception or design of the work: Claudia Gabriele Werner
- Data collection: Claudia Gabriele Werner
- Data analysis and interpretation: Claudia Gabriele Werner
- Drafting the article: Claudia Gabriele Werner
- Critical revision of the article: Frank-Dietrich Wagner
- Final approval of the version to be published: Claudia Gabriele Werner, Frank-Dietrich Wagner
-

Acknowledgements: We thank Frank-Dietrich Wagner for his thoroughly review of the manuscript.

8. References

1. Wu D, Chen W. (2024) Molecular mechanisms and emerging therapies in wild-type transthyretin amyloid cardiomyopathy. Heart Fail Rev. 29(2): 511-521.
2. Ando Y, Coelho T, Berk JL, et al. (2013) Guideline of transthyretin-related hereditary amyloidosis for clinicians. Orphanet J Rare Dis. 8: 31.
3. Rowczenio DM, Noor I, Gillmore JD, et al. (2014) Online registry for mutations in hereditary amyloidosis including nomenclature recommendations. Hum Mutat. 35: E2403-E2412.
4. Damy T, Kristen AV, Suhr OB, et al. (2019) Transthyretin cardiac amyloidosis in continental western Europe: An insight through the transthyretin amyloidosis outcomes survey (thaos). Eur Heart J. 43: 391-400.
5. Zhou J, Li Y, Geng J, et al. (2023) Recent progress in the development and clinical application of new drugs for transthyretin cardiac amyloidosis. J Cardiovasc Pharmacol. 82: 427-437.
6. Delgado D, Dabbous F, Shivappa N, et al. (2025) Epidemiology of transthyretin (ATTR) amyloidosis: A systematic literature review. Orphanet Journal of Rare Diseases. 20: 29.
7. Bruno M, Castaño A, Burton A, et al. (2021) Transthyretin amyloid cardiomyopathy in women: Frequency, characteristics, and diagnostic challenges. Heart Fail Rev. 26(1): 35-45.
8. (2025) Union Register of medicinal products. Website: https://ec.europa.eu/health/documents/community-register/html/h717.htm
9. Kazi DS, Bellows BK, Baron SJ, et al. (2020) Cost-effectiveness of tafamidis therapy for transthyretin amyloid cardiomyopathy. Circulation. 141: 1214-1224.
10. González-López E, Gallego-Delgado M, Guzzo-Merello G, et al. (2015) Wild-type transthyretin amyloidosis as a cause of heart failure with preserved ejection fraction. Eur Heart J. 36: 2585-2594.
11. Hahn VS, Yanek LR, Vaishnav J, et al. (2020) Endomyocardial Biopsy Characterization of Heart Failure with Preserved Ejection Fraction and Prevalence of Cardiac Amyloidosis. JACC Heart Fail. 8(9): 712-724.
12. Lindmark K, Pilebro B, Sundström T, et al. (2021) Prevalence of wild type transtyrethin cardiac amyloidosis in a heart failure clinic. ESC Heart Fail. 8(1): 745-749.
13. Jerome S, Farrell MB, Warren J, et al. (2023) Cardiac Amyloidosis Imaging, Part 3: Interpretation, Diagnosis, and Treatment. J Nucl Med Technol. 51: 102-116.
14. Singh V, Falk R, Di Carli MF, et al. (2019) State-of–art radionuclide imaging in cardiac transthyretin amyloidosis. J Nucl Cardiol. 26: 158-173.
15. Garcia-Pavia P, Benge F, Brito D, et al. (2021) Expert consensus on the monitoring of transthyretin amyloid cardiomyopathy. European Journal of Heart Failure. 23: 895-905.
16. Maurer MS, Schwartz JH, Gundapaneni B, et al. (2018) Tafamidis treatment for patients with transthyretin amyloid cardiomyopathy. N Engl J Med. 379(11): 1007-1016.
17. Damy T, Angeli FS, Wang R, et al. (2024) Long-term all-cause and cardiovascular-related mortality in patients with ATTR-CM treated with continuous tafamidis vs placebo to tafamidis by NAC stage at baseline. Eur Heart J. 45.
18. Wu F, Zhu He, Zhang Y. (2024) Analysis of post-market adverse events of tafamidis based on the FDA adverse event reporting system. Sci Rep. 14(1): 13691.
19. ESC CHF Guidelines. (2021) 2021 ESC guidelines for the diagnosis and treatment of acute and chronic heart failure. ESC Task force. Eur Heart J. 42: 3599-3726.
20. Heidenreich PA, Bozkurt B, Aguilar D, et al. (2022) 2022 AHA/ACC/HFSA guideline for the management of heart failure: a report of the American College of Cardiology/American Heart Association Joint Committee on Clinical Practice Guidelines. J Am Coll Cardiol. 79: e263-e421.
21. Kittleson MM, Ruberg FL, Ambardekar AV, et al. (2023) 2023 ACC Expert Consensus Decision Pathway on Cardiac Amyloidosis. JACC. 81(11): 1076-1126.
22. Hammarström P, Jiang X, Hurshman AR, et al. (2002) Sequence-dependent denaturation energetics: A major determinant in amyloid disease diversity. Proc Natl Acad Sci USA. 99: 16427-16432.
23. Ji A, Wong P, Judge DP, et al. (2023) Acoramidis produces near-complete TTR stabilization in blood samples from patients with variant transthyretin amyloidosis that is greater than that achieved with tafamidis. Eur Heart J. 44.
24. Judge DP, Heitner SB, Falk RH, et al. (2019) Transthyretin stabilization by AG10 in symptomatic transthyretin cardiomyopathy. J Am Coll Cardiol. 74(3): 285-295.

25. Gillmore JD, Judge DP, Cappelli F, et al. (2024) Efficacy and safety of acoramidis in transthyretin amyloid cardiomyopathy. N Engl J Med. 390: 132-142.
26. Judge DP, Alexander KM, Cappelli F, et al. (2025) Efficacy of Acoramidis on All-Cause Mortality and Cardiovascular Hospitalization in Transthyretin Amyloid Cardiomyopathy. J Am Coll Cardiol. 85(10):1003-1014.
27. Judge DP, Gillmore JD, Alexander KM, et al. (2025) Long-Term Efficacy and Safety of Acoramidis in ATTR-CM: Initial Report from the Open-Label Extension of the ATTRibute-CM Trial. Circulation. 151(9): 601-611.
28. Habtemariam BA, Karsten V, Attarwala H, et al. (2021) Single-dose pharmacokinetics and pharmacodynamics of transthyretin targeting N-acetylgalactosamine-small interfering ribonucleic acid conjugate, vutrisiran, in healthy subjects. Clin Pharmacol Ther. 109: 372-382.
29. Alnylam Pharmaceuticals. (2025) Prescribing information: AMVUTTRA® (vutrisiran) injection, for subcutaneous use.
30. Fontana. (2024) Vutrisiran HELIOS-B trial.
31. Keller DM, Straburzyńska-Migaj E, Lesiak M. (2025) Advancing treatments for transthyretin amyloid cardiomyopathy: Innovations in RNA silencing, gene editing, TTR stabilization, and degradation. Pol Heart J. 83(2): 121-137.
32. Murad A, Lasko MJ, Badri P, et al. (2024) A Phase 1, Single Ascending Dose Study to Evaluate ALN-TTRsc04, a Next-Generation RNA Interference Therapeutic, in Healthy Participants for Potential Treatment of Transthyretin Amyloidosis. American Heart Association Scientific Sessions.
33. Van der Meer PV, Lairez O, Donal E, et al. (2024) Long-term safety and efficacy of antibody ALXN2220 for depletion of cardiac amyloid transthyretin: Results of treatment beyond 12 months in the open-label extension of study NI006-101. Eur Heart J. 45.
34. Gillmore JD, Gane E, Taubel J, et al. (2021) CRISPR-Cas9 *in vivo* gene editing for transthyretin amyloidosis. N Engl J Med. 385(6): 493-502.
35. Press Release Intellia Therapeutics. (2025) Intellia therapeutics announces first patient dosed in the magnitude-2 phase 3 study of Nexiguran Ziclumeran (NEX-Z), A one-time gene editing-based treatment for Transthyretin (ATTR) Amyloidosis with Polyneuropathy - Intellia therapeutics.
36. Zhang X, Goel V, Attarwala H, et al. (2020) Patisiran pharmacokinetics, pharmacodynamics, and exposure-response analyses in the phase 3 APOLLO trial in patients with hereditary transthyretin-mediated (hATTR) amyloidosis. J Clin Pharmacol. 60(1): 37-49.
37. Maurer MS, Kale P, Fontana M, et al. (2023) Patisiran treatment in patients with transthyretin cardiac amyloidosis. N Engl J Med. 389(17): 1553-1565.
38. Wall JS, Selvarajah S, Foster JS, et al. (2023) Characterization of AT-02, a pan-amyloid-binding peptide fusion immunoglobulin with high binding potency, complement activation, and immune cell stimulation. Eur Heart J. 44.
39. Wall J, Klein M, Guthrie S, et al. (2024) The peptide fusion immunoglobulin, AT-02, exhibits highly potent pan-amyloid reactivity and immunomodulation. J Card Fail. 30(1): 210.
40. Fontana M, Buchholtz K, Engelmann M, et al. (2022) NNC6019-0001, a humanized monoclonal antibody, in patients with transthyretin amyloid cardiomyopathy (ATTR-CM): Rationale and study design of a phase 2, randomized, placebo-controlled trial. Eur Heart J. 43.
41. Suhr OB, Grogan M, Silva AMD, et al. (2024) PRX004 in variant amyloid transthyretin (ATTRv) amyloidosis: results of a phase 1, open-label, dose-escalation study. Amyloid. 1-8.
42. Pacher P, Steffens S, Haskó G, et al. (2018) Cardiovascular effects of marijuana and synthetic cannabinoids: the good, the bad, and the ugly. Nat. Rev. Cardiol.15: 151-166.
43. Ibsen MS, Connor M, Glass M. (2017) Cannabinoid CB1 and CB2 receptor signaling and bias. Cannabis Cannabinoid Res. 2: 48-60.
44. Wei TT, Chandy M, Nishiga M, et al. (2022) Cannabinoid receptor 1 antagonist genistein attenuates marijuana-induced vascular inflammation. Cell. 185(10): 1676-1693.
45. Press Release YolTech; March11, 2025: Yoltech Therapeutics Announces Positive Clinical Results For Yolt-201, An Investigational In Vivo Gene Editing Therapy For Transthyretin Amyloid Cardiomyopathy (Attr-Cm)-Press Release-Yoltech.
46. Sinha S, Du Z, Maiti Pet al. (2012) Comparison of three amyloid assembly inhibitors: the sugar scyllo-inositol, the polyphenol epigallocatechin gallate, and the molecular tweezer CLR01. ACS Chem Neurosci. 3: 451-458.
47. Attar A, Chan WT, Klarner FG, et al. (2014) Safety and pharmacological characterization of the molecular tweezer CLR01-a broad-spectrum inhibitor of amyloid proteins' toxicity. BMC Pharmacol Toxicol. 15: 23.
48. Hanson JLS, Arvanitis M, Koch CM, et al. (2018) Use of serum transthyretin as a prognostic indicator and predictor of outcome in cardiac amyloid disease associated with wild-type transthyretin. Circ Heart Fail. 11(2): e004000.
49. Siddiqi OK, Mints YY, Berk JL et al. (2022) Diflunisal treatment is associated with improved survival for patients with early stage wild-type transthyretin (ATTR) amyloid cardiomyopathy: the Boston University Amyloidosis Center experience. Amyloid. June; 29(2): 71-78.
50. Cardoso I, Martins D, Ribeiro T, et al. (2010) Synergy of combined doxycycline/TUDCA treatment in lowering transthyretin deposition and associated biomarkers: Studies in FAP mouse models. J Transl Med. 8: 74.
51. Wixner J, Pilebro B, Lundgren HE, et al. (017) Effect of doxycycline and ursodeoxycholic acid on transthyretin amyloidosis. Amyloid. 2(24): 78-79.
52. Ferreira N, Saraiva MJ, Almeida MR. (2011) Natural polyphenols inhibit different steps of the process of transthyretin (TTR) amyloid fibril formation. FEBS Lett. 585(15): 2424-2430.
53. Aus Dem Siepen F, Bauer R, Aurich M, et al. (2015) Green tea extract as a treatment for patients with wild-type transthyretin amyloidosis: An observational study. Drug Des Devel Ther. 9: 6319-6325.
54. Saelices L, Nguyen BA, Chung K et al. (2019) A pair of peptides inhibits seeding of the hormone transporter transthyretin into amyloid fibrils. J. Biol. Chem. 294(15): 6130-6141.
55. Saelices L, Pokrzywa M, Pawelek K, et al. (2018) Assessment of the effects of transthyretin peptide inhibitors in Drosophila models of neuropathic ATTR. Neurobiol. Dis. 120: 118-125.

9. Appendix

Appendix 1: Clinical characteristics of ATTRv versus ATTRwt.

	Age at Symptom onset (y)	Percentage of Men (%)	Underlying Condition	Duration of Symptoms before Diagnosis (y)	Median Survival Time after Diagnosis (y)	Organs or Systems Involved
ATTRv	> 20	70-80	TTR mutation	2.8-4.3	2.5-3.5 (when hear is involved); 8-10 (when only nerves are involved)	Hear, peripheral nerves, kidney, eyes (vitreous opacity)
ATTRwt	> 60	85-90	Age	~2	3.6-4.8	Hear, muscle (carpal tunnel syndrome, conical stenosis, and biceps rupture), occasional peripheral nerve

ATTRv, Genetic/mutant transthyretin amyloidosis; ATTRwt, wild-type transthyretin amyloidosis.
Source: Figure adopted from Zhou, et al. 2023 [5].

Appendix 2: Overview of approved Disease Modifying Therapie's for ATTR-CM in the EU and USA.

ATTR-CM Therapies	Mechanism of action	FDA/EMA approval	Dosing Regimen	Costs per month
TTR-stabilizer				
Tafamidis	TTR stabilizer: Tafamidis binds to the thyroxine binding sites of TTR, thereby stabilizing the tetramer and slowing down the cleavage into monomers, the rate-determining step in the amyloidogenic process.	FDA 2019 (VYNDAMAX™ from Pfizer), EMA 2020 (VYNDAQEL® from Pfizer)	Tafamidis 61 mg (bioequivalent to tafamidis meglumine 80 mg) per os once daily	US$22,000 (VYNDAMAX™) 11.778,41 € (VYNDAQEL®)
Acoramidis	Next generation TTR stabilizer: Acoramidis forms hydrogen bonds with neighboring serine residues within both thyroxine binding sites of the tetramer (mimicking the mode of action of the protective T119M variant). This interaction stabilizes the tetramer and slows down the cleavage into monomers, the rate-determining step in the amyloidogenic process.	FDA 2024 (ATTRUBY™ from BridgeBio), EMA 2025 (BEYONTTRA® from Bayer)	Acoramidis 712 mg per os twice daily (total daily dose of 1 424 mg)	US$20,036 (ATTRUBY™) 11.695,68€ (BEYONTTRA®)
TTR Gene silencer				
Vutrisiran	Next generation siRNA: Vutrisiran is a double-stranded siRNA-GalNAc conjugate that causes degradation of mutant and wild-type TTR mRNA through RNA interference, which results in a reduction of serum TTR protein and TTR protein deposits in tissues.	FDA March 2025 (AMVUTTRA™ from Alnylam) EMA June 2025 (AMVUTTRA® from Alnylam)	Vutrisiran 25 mg subcutaneously once every 3 months	US$39,666 (AMVUTTRA™) 26.599,01€ (AMVUTTRA®)

Abbreviations: GalNAc = N-acetylgalactosamine, siRNA = small interfering RNA, TTR = transthyretin,
Source: Based on data reported in Prescribing Information of VYNDAMAX™ (10/2023), VYNDAQEL® (02/2023), ATTRUBY™ (11/2024), BEYONTTRA® (06/2025), AMVUTTRA™ (03/2025) and (AMVUTTRA® (06/2025).

Appendix 3: ATTR-CM - current drug pipeline.

Product (Sponsor)	Mechanism of Action	Route of administration/ Dosing frequency	Primary Outcome Measure(s)	NCT (clinicaltrials.gov)
Phase 3				
Eplontersen (Ionis/Astra Zeneca)	Ligand-conjugated ASO medicine designed to inhibit the production of TTR protein	Eplontersen given as SC injection once every 4 weeks	Composite Outcome of cardiovascular (CV) mortality and recurrent CV clinical events from Baseline up to Week 140	**NCT04136171** (CARDIO-TTRansform):Active, not recruiting, SCE: 08/2026
Nucresiran (Alnylam Pharmaceuticals)	Third generation (long acting) siRNA based on IKARIATM platform methodology: Degradation of mutant and wild-type TTR mRNA resulting in a reduction of serum TTR protein and amyloid deposits in tissues.	Nucresiran is given as SC injection once every 6 months	Composite Outcome of cardiovascular (CV) mortality and recurrent CV clinical events from baseline to end of double-blind treatment period (estimated 32 months, maximum 5 years)	**NCT07052903** (TRITON-CM): Recruiting, SCE: 11/2032
ALXN2220 (Alexion Pharmac.)	Human anti-ATTR mAb designed to deplete cardiac ATTR deposits (ATTR amyloid depleter)	IV infusion every 4 weeks for at least 24 months up to a max. of 48 months	Composite Outcome of all-cause mortality and cardiovascular clinical events up to 48 months	**NCT06183931** (DepleTTR-CM): Active, not recruiting, SCE: 10/2027
Nexiguran-ziclumeran (Intellia Therap.)	Gene-editing using CRISP/Cas9 technology to inactivate the *TTR* gene that encodes for the mutated TTR protein.	Single infusion as a one-time treatment	Composite outcome of cardiovascular mortality and cardiovascular events over 18 months and (depending on event rates) up to 48 months.	**NCT06128629** (MAGNITUDE): Recruiting, SCE: 04/2028
Phase 2				
AT-02 (Attralus Inc.)	Humanized IgG1 mAb fused to the pan-amyloid binding peptide p5R. AT-02 promotes macrophage mediated phagocytosis of all types of amyloid deposits (pa-ATTR amyloid depleter).	Intravenous infusion once every 2 or 4 weeks for 104 weeks	Safety and tolerability of AT-02 through detection of TEAEs and change in clinical laboratory parameters from baseline up to 112 weeks	**NCT05951049** (OLE trial of NCT05521022): Recruiting, SCE: 02/2026
Coramitug (Novo Nordisk)	Human anti-ATTR mAb designed to deplete cardiac ATTR deposits through antibody-mediated phagocytosis (ATTR amyloid depleter)	IV-infusion every 4 weeks until week 52 IV infusion every 4 weeks until week 140	At week 52: change in 6MWT distance and NTproBNP level Number of TEAEs from baseline up to week 156	**NCT05442047:** Completed 05/2025 **NCT06260709:** Recruiting, SCE: 08/2028
Genistein (London Health Sciences Centre Research Institute)	Soybean isoflavone with anti-inflammatory and antioxidant properties, cannabinoid receptor 1 antagonist in the vasculature	Ascending oral doses of genistein (up to 750 mg BD) over 12 weeks	Safety profile and effect of genistein on inflammatory, cardiac biomarkers, Complete metabolic profile, 6MWT, echocardiography	**NCT06634108** (GASPAR): Recruiting, SCE: 12/2025
Phase 1				
YOLT-201 (YolTech)	Gene-editing drug using CRISP/Cas9 technology to inactivate the TTR gene that encodes for the mutated TTR protein.	Single infusion as a one-time treatment	Safety, tolerability and preliminary efficacy (6MWT, KCCQ, cardiac biomarkers, echocardiography, cardiac MRI) up to 1 year	**NCT06082050:** Recruiting, SCE: 02/2026

Abbreviations: ASO = antisense oligonucleotide, IV = intravenous, KCCQ, = Kansas City Cardiomyopathy Questionnaire, MRI = magnet resonance imaging, mAb= monoclonal antibody, 6MWT = 6-minute walk test, N-terminal-pro brain natriuretic peptide = NTproBNP, OLE = Open Label Extension, SC = subcutaneous, SCE = study completion (estimated), TEAE = Treatment Emergent Adverse Event, TTR = transthyretin

Source: Based on data at CLINICALTRIALS.GOV (latest access at 30st July 2025).