NON-INTERVENTIONAL (NI) STUDY PROTOCOL

Study information

Title	Patient outcomes and clinical characteristics of ATTR-CM patients treated with tafamidis: A single center experience in Turkey- Study ATTREAL
Protocol number	B3461126
Protocol version identifier	2.0
Date	16 December 2025
EU Post Authorization Study (PAS) register number	EUPAS1000000561
Active substance	N07XX08
Medicinal product	Tafamidis
Research question and objectives	The aim of this study is to describe the clinical characteristics and outcomes of patients with ATTR amyloid cardiomyopathy (ATTR-CM) treated with tafamidis at a single center in Turkey. Primary Objective: To describe the rate of hospitalizations and emergency admissions due to chronic heart failure in the patients treated with Tafamidis Secondary Objectives: - To describe the rate of cardiovascular-related hospitalizations and overall survival among patients treated with Tafamidis during Tafamidis exposure time period. - To describe the presence of cardiac and extracardiac red flags (Extra-cardiac Red Flags: Carpal Tunnel Syndrome, Peripheric Neuropathy, Lumbar Stenosis, Renal Insufficiency, Cardiac Red Flags: Elevated NT Pro BNP and troponin, Pseudo infarct Sign, Voltage Mismatch, Conduction Disorder, Increased Wall Thickness (Interventricular septum), Pericardial Effusion)) in patients with ATTR - CM treated with tafamidis during the baseline period prior to initiation of tafamidis treatment.

	- To describe the clinical and functional outcomes in ATTR CM patients during the baseline period prior to tafamidis treatment and during the follow-up period of treatment with tafamidis such as HRQoL (Health-Related Quality of Life) measures, concomitant medication, biomarkers (NT pro-BNP, troponin), echocardiographic parameters (6 min walk test, Kansas City Cardiomyopathy Questionnaire Score, Creatinine eGFR, Ejection Fraction, Interventricular Septum, E/A ratio, E/e' mean ratio, Tricuspid Regurgitation Velocity, Tricuspid Annular Plane Systolic Excursion).
Country(ies) of study	Turkey
Author	Furkan Erdoğan, MD, [redacted]

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1. LIST OF ABBREVIATIONS

Abbreviation	Definition
AEM	Adverse Event Management
AL	Amyloid Light Chain (other type of amyloidosis to differentiate from ATTR)
ALT	Alanine Transaminase
AF	Atrial Fibrillation
AST	Aspartate Aminotransferase
ATTR-ACT	TTR Amyloidosis Cardiomyopathy Clinical Trial
ATTR CM	TTR amyloid cardiomyopathy
AV	Atrioventricular
BMI	Body Mass Index
BPM	Beats Per Minute
CA	Cardiac Amyloidosis
CIOMS	Council for International Organizations of Medical Sciences
CSA	Clinical Study Agreement
CHF	Chronic Heart Failure
eCRF	Electronic Case Report Form
CRO	Clinical Research Organization
COPD	Chronic Obstructive Pulmonary Disease
CV	Cardiovascular
E/A Ratio	Early to Atrial filling velocity ratio
EC	Ethics Committee
ECG	Electrocardiogram
ECHO	Echocardiography
EDP	Exposure during pregnancy
eGFR	Estimated Glomerular Filtration Rate
E/e'	Early mitral inflow velocity and mitral annular early diastolic velocity ration
EF	Ejection Fraction
EHR	Electronic Health Record
EMA	European Medicines Agency
ENCePP	European Network of Centres for Pharmacoepidemiology and Pharmacovigilance
FDA	Food and Drug Administration
GPP	Good Pharmacoepidemiology Practices
HF	Heart Failure (symptom management focus)
HFpEF	Heart Failure with Preserved Ejection Fraction
HFmrEF	Heart Failure with mildly reduced ejection fraction
HFrEF	Heart Failure with reduced ejection fraction
HRQoL	Health-Related Quality of Life
hATTR	Hereditary TTR Amyloidosis
ICD	International Classification of Diseases

ICMJE	International Committee of Medical Journal Editors (ICMJE)
IRB	Institutional Review Board
ISPOR-ISPE	International Society for Pharmacoeconomics and Outcomes Research - International Society for Pharmaceutical Engineering
IVS	Interventricular Septum
KCCQ	Kansas City Cardiomyopathy Questionnaire
LAVI	Left Atrial Volume Index
LVMI	Left Ventricular Mass Index
LVEDD	Left Ventricular End-Diastolic Dimension
LVESD	Left Ventricular End-Systolic Dimension
MRI	Magnetic Resonance Imaging
NIS	Non-interventional study
NT-proBNP	N-terminal pro-B-type Natriuretic Peptide
NYHA class	The NYHA (New York Heart Association) Functional Classification
PW	Posterior Wall
RWD	Real World Data
PYP/ DPD/ HMDP	Pyrophosphate / Diphosphono-1,2-propanodicarboxylic acid / Hydroxymethylene Diphosphonat
RBP	Retinol-binding protein
RWE	Real-World Evidence
AEs/SAEs	Adverse Events (AEs) and Serious Adverse Events (SAEs)
SAP	Statistical Analysis Plan
SAVR	Surgical Aortic Valve Replacement
SOP	Standard Operating Procedures
sPAP	Systolic Pulmonary Artery Pressure
SPECT-CT	Single-Photon Emission Computed Tomography
T4	Thyroxine
T60A	Genetic mutation where the amino acid threonine (T) at position 60 of a protein is replaced by alanine (A)
TAPSE	Tricuspid Annular Plane Systolic Excursion
TAVI	Transcatheter Aortic Valve Implantation
TTR	Transthyretin (protein involved in amyloidosis)
TTR Stabilizers	Transthyretin Stabilizers
V122I	Genetic mutation where amino acid valine (V) is substituted by isoleucine (I) at position 122
wtATTR	Wild-Type TTR Amyloidosis
YRR	Your Reporting Responsibilities
6MWT	6-Minute Walk Test
99mTc	Technetium-99m (used in imaging studies)

2. RESPONSIBLE PARTIES

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3. ABSTRACT

Title: Patient outcomes and clinical characteristics of ATTR-CM patients treated with tafamidis: A single center experience in Turkey

Version 1.0, 16 December 2025

Furkan Erdoğan, [redacted]

Rationale and background: ATTR-CM is an advanced and complicated disease caused by the aggregation of misfolded TTR proteins in the heart, resulting in restrictive cardiomyopathy and heart failure. Tafamidis, an FDA approved agent, TTR stabilizer, has been studied for its potential to modify the disease and improve clinical outcomes in patients with wild-type or variant transthyretin amyloidosis. Although tafamidis has changed the treatment algorithm, patient outcomes and clinical characteristics in the real-world settings, especially in populations with unique genetic, clinical, and healthcare system characteristics such as Turkey is poorly understood. Regional differences in genetic mutations, access to advanced diagnostic tools, and healthcare practices may influence patient outcomes. Tafamidis is currently the only approved treatment option for ATTR-CM, it is approved and reimbursed for patients in Turkey.

This study represents the first comprehensive descriptive study of patient outcomes following tafamidis treatment in Turkey. The findings will enhance understanding of ATTR-CM epidemiology and serve as a valuable resource for improving disease awareness among cardiologists and shaping future diagnostic and therapeutic guidelines.

Research question and objectives: The aim of this study is to describe the clinical characteristics and outcomes of patients with ATTR amyloid cardiomyopathy (ATTR-CM) treated with tafamidis at a single center in Turkey.

Primary Objective: to describe the rate of hospitalizations and emergency admissions due to chronic heart failure in the patients treated with Tafamidis.

Secondary Objectives:

- To describe the rate of cardiovascular-related hospitalizations and overall survival among patients treated with Tafamidis during Tafamidis exposure time period.
- To describe the presence of cardiac and extracardiac red flags (Extra-cardiac Red Flags: Carpal Tunnel Syndrome, Peripheric Neuropathy, Lumbar Stenosis, Renal Insufficiency, Cardiac Red Flags: Elevated NT Pro BNP and troponin, Pseudo infarct Sign, Voltage Mismatch, Conduction Disorder, Increased Wall Thickness (Interventricular septum), Pericardial Effusion)) in patients with ATTR - CM treated with tafamidis during the baseline period prior to initiation of tafamidis treatment.
- To describe the clinical and functional outcomes in ATTR CM patients during the baseline period prior to tafamidis treatment and during the follow-up period of treatment with tafamidis such as HRQoL (Health-Related Quality of Life) measures, concomitant medication, biomarkers (NT pro-BNP, troponin), echocardiographic parameters (6 min walk test, Kansas City Cardiomyopathy Questionnaire Score, Creatinine eGFR, Ejection Fraction, Interventricular Septum, E/A ratio, E/e' mean ratio, Tricuspid Regurgitation Velocity, Tricuspid Annular Plane Systolic Excursion).

Study design: This is a single-center, non-interventional, retrospective cohort study.

Population: Patients must meet the following inclusion criteria: 1) >18 years old at the time of diagnosis; ii) diagnosed with hereditary or wild type ATTR-CM confirmed non-invasively (Tc99m PYP bone scintigraphy and/or SPECT-CT) in Samsun Education and Research Hospital – Cardiology Outpatient Clinic between January 2023 – January 2025; iii) currently receiving or who have received tafamidis treatment for ATTR-CM; iv) Availability of complete medical records, including baseline clinical characteristics, treatment outcomes, and follow-up data from electronic health record systems of hospital or physical patient files. Patients will be excluded if participated in studies involving investigational drugs within 24 months before the index date.

Variables: Demographics**,** NYHA class, 6-minute walk test (6MWT), quality of life, Electrocardiogram (ECG), echocardiography, and Magnetic Resonance Imaging (MRI) data, blood parameters, including troponin and N-terminal pro Brain Natriuretic Peptide (NT pro-BNP), Estimated Glomerular Filtration Rate (eGFR) Biomarkers, Bone scintigraphy/SPECT Imaging, Genetic tests, Serum protein electrophoresis, Serum/urine immunofixation electrophoresis

Data source: Pseudonymized data will be entered in CRF from individual patient medical records maintained in a single center in Turkey according to clinical practice. Data will be collected using electronic case report form (eCRF).

Sample size: Due to the absence of a priori hypothesis, sample size calculations are not applicable, and no formal sample size calculation has been performed. According to the descriptive nature of the study, we have planned a sample of around 70 subjects approximating 10% of the entire population diagnosed with ATTR-CM in Turkey, with no formal calculation related to a specific endpoint.

Data analysis: Descriptive statistics will be reported and no effort to determine statistical significance will be made.

Milestones:

Milestone	Planned Date
Registration in the HMA-EMA Catalogues of RWD studies	30 July 2025
Start of data collection	16 October 2025
End of data collection	30 June 2026
Final study report	31 December 2026

4. AMENDMENTS AND UPDATES

Version Identifier	Date	Amendment Type (substantial or administrative)	Protocol Section(s) Changed	Summary of Amendment(s)	Reason
2.0	16 Dec 2025	Substantial	5. Milestones	Start of data collection, end of data collection, and final study report dates are postponed per the actual data collection start date.	Completion of site agreement and therefore the SIV were completed later than initially planned. Due to these, data collection has started later than initially planned and actual data collection start date is mentioned in the revised version. Due to the increase in subject number, longer duration is now anticipated for completion of data entry. Therefore, dates of end of data collection and final study report are also postponed accordingly.
		Substantial	3. Abstract		
		Substantial	8.5. Study Size	Sample size is increased to 70.	Since the start of data collection is postponed, number of eligible patients at the study site has increased during that duration. To obtain more comprehensive data, it is decided to include all eligible patients to the study.
		Substantial	3. Abstract		
		Substantial	8.2. Setting		
		Administrative	8.3. Variables	Variable name “high-sensitivity troponin” is changed to “ troponin”.	Troponin assay type is changed to be compatible with the study site’s assay type.
		Administrative	8.3. Variables	Assessment period of the variable “NYHA Class” is corrected to include the “follow-up	Due to an error and oversight, “follow-up assessment period” of the “NYHA Class” was initially omitted and now updated accordingly.

Version Identifier	Date	Amendment Type (substantial or administrative)	Protocol Section(s) Changed	Summary of Amendment(s)	Reason
				assessment period".	
		Administrative	8.3. Variables	Operational definition of the variable "Carpal Tunnel Syndrome" is changed to "History of Carpal Tunnel Syndrome (yes/no/missing) as documented in the source data".	Due to an error and oversight, operational definition of the "Carpal Tunnel Syndrome" was initially mentioned incorrectly and now updated accordingly.
		Administrative	8.3. Variables	Variable name "Tafamidis treatment start date" is changed to "Tafamidis treatment prescription date". Operational definition of the same variable is also updated accordingly.	Due to an error and oversight, variable name and operational definition of the "Tafamidis treatment prescription date" were initially mentioned incorrectly and now updated accordingly.
		Administrative	Secondary Objective	Patients' follow-up period is clarified.	Since the follow-up duration of the patients will change (approximately min. 6 months to max. 30 months) depending on the Tafamidis exposure time period, the duration of follow-up period of approximately 12 months was removed from the formulation of the secondary objective.

Version Identifier	Date	Amendment Type (substantial or administrative)	Protocol Section(s) Changed	Summary of Amendment(s)	Reason
					Individual patient-level follow-up will be used.
		Administrative	8.2. Setting	Cut-off/end of the patients' follow-up period is clarified.	Study team decided to clarify the cut-off/end of the patients' follow-up period and the relevant paragraph is included in the 8.2 Setting section.
		Administrative	8.6. Data Management	Cut-off/end of the patients' follow-up period is clarified.	"Data Collection and Entry" section of the 8.6. Data Management is revised to reflect the current data collection period.
		Administrative	8.3. Variables	Units of sodium, potassium, albumin, NT pro-BNP, and serum free light chain levels (kappa and lambda) are changed.	Units of some laboratory parameters are changed to align with the actual units implemented by the site.
		Administrative	Overall	Minor clerical errors were corrected.	

5. MILESTONES

Milestone	Planned Date
Registration in the HMA-EMA Catalogues of RWD studies	30 July 2025
Start of data collection	16 October 2025
End of data collection	30 June 2026
Final study report	31 December 2026

6. RATIONALE AND BACKGROUND

Amyloidosis is a systemic disorder characterized by the abnormal extracellular deposition of misfolded proteins, which aggregate into amyloid fibrils. These fibrils can deposit in various tissues and organs, impairing their normal function. The two most common forms of cardiac amyloidosis are AL amyloidosis and ATTR amyloidosis, each caused by different proteins.[1-3]

ATTR amyloidosis involves the deposition of misfolded TTR, a protein primarily produced by the liver. ATTR amyloidosis can be hereditary or wild type. Hereditary ATTR amyloidosis (hATTR) is associated with mutations in the TTR gene, while wild-type ATTR (wtATTR) occurs due to aging and is more common in older individuals. [2-3]

Treatment strategies aim to reduce amyloid deposition and manage the associated heart failure symptoms. In ATTR amyloidosis, tafamidis has shown efficacy in reducing disease progression and improving clinical outcomes, including heart-related symptoms.[4]

Cardiac amyloidosis is characterized by the extracellular accumulation of abnormal proteins. Regardless of the subtype of the deposited amyloid, the disease results in progressive heart failure if left untreated. TTR is a tetrameric transport protein primarily synthesized in the liver, responsible for carrying T4 and RBP. Under pathological conditions, TTR dissociates into monomers, which misfold and aggregate into insoluble amyloid fibrils. These fibrils infiltrate the cardiac interstitium, causing myocardial stiffening, diastolic dysfunction, and eventually heart failure.[5]

1. **wtATTR**: Studies show that approximately 25% of individuals over 80 years have cardiac amyloid deposits.[6] Occurs due to age-related destabilization of TTR, typically presenting after the age of 60–65 years More prevalent in men and often associated with carpal tunnel syndrome and spinal stenosis as early non-cardiac manifestations**.** Historically underdiagnosed but increasingly recognized with advancements in imaging techniques.[7]
2. **hATTR:** The prevalence is influenced by regional genetic factors. Caused by autosomal dominant mutations in the TTR gene. Over 120 mutations have been identified, with the most common being V122I (3-4% of African Americans) and T60A (Northern Europe).[8,9]

Patients with ATTR-CM typically present with symptoms of heart failure, including dyspnea, fatigue, and peripheral edema. Key distinguishing features include:

- Preserved ejection fraction (HFpEF): ATTRwt is an underdiagnosed disease that accounts for a significant number (13%) of HFpEF cases.[10] ATTR-CM acts like heart failure makes diagnosis challenging with preserved ejection fraction due to restrictive cardiomyopathy but often progresses despite standard HF therapies. [11]
- Conduction system disease: Manifesting as arrhythmias or atrioventricular block, necessitating pacemaker implantation. [12]
- Red flag signs: Biventricular thickening without history of hypertension, low-voltage ECG despite hypertrophy, atrial fibrillation or AV block requiring pacemaker implantation and unexplained heart failure with HFpEF. [13]

The diagnostic work-up for CA begins with the identification of clinical features, ECG and imaging findings suggestive or compatible with CA, and often requires the histological demonstration of amyloid deposition, except when Tc99m PYP/DPD/HMDP scintigraphy shows an intense myocardial uptake (Perugini scores 2-3) in the absence of a monoclonal gammopathy. [14]

ATTR-CM was historically diagnosed with endomyocardial biopsy, which is invasive and carries risks. Recent advances in non-invasive diagnostic methods, including technetium-99m pyrophosphate (99mTc-PYP) scintigraphy, cardiac MRI, and serum biomarkers like NT-proBNP, have revolutionized early detection. [14,15]

Tafamidis Treatment

Tafamidis is a first-in-class TTR stabilizer that prevents TTR tetramer dissociation, thereby halting the amyloidogenic cascade. The landmark ATTR-ACT trial demonstrated that tafamidis significantly reduced all-cause mortality, cardiovascular hospitalizations, and functional decline in patients with ATTR-CM.[4]

ATTR-ACT trial, a pivotal multinational, double-blind, placebo-controlled phase III study demonstrated the efficacy of tafamidis in significantly reducing both all-cause mortality and cardiovascular hospitalizations. 441 patients with ATTR-CM were randomly assigned in a 2:1:2 ratio to receive tafamidis meglumine at doses of 80 mg or 20 mg, or placebo, once daily for 30 months. Endpoints resulted as reducing mortality (Relative risk reduction of 30% (P<0.001) and the rate of hospitalizations. (lower in the tafamidis group (0.48 per year) compared to the placebo group (0.70 per year; P<0.001). Improvements were consistent both for wild-type and hereditary ATTR-CM. Both tafamidis doses (80 mg and 20 mg) were effective, with similar outcomes. Patients treated with tafamidis demonstrated a smaller decline in the 6MWT distance and better maintenance of functional capacity compared to placebo. Quality of life, assessed using the KCCQ, showed significantly less deterioration in the tafamidis group. This study established tafamidis as the first FDA-approved therapy for this condition.[4]

[redacted]

Pablo Garcia-Pavia et al. observed that patients treated with tafamidis experienced improved survival rates compared to those not receiving the treatment survival in a Real-World Cohort of Patients with ATTR. [16] Another Real-World data from a Tertiary Center's research reported that tafamidis treatment led to reductions in cardiovascular hospitalizations and enhanced survival among ATTR-CM patients.[17]

Hussain K et al. evaluated the impact of tafamidis on overall survival in patients with ATTR-CM within a community-based cohort in a retrospective observational study analyzing patient data from a community hospital comparable to the current setting. The study supports the efficacy of tafamidis observed in clinical trials, indicating its beneficial impact extends to routine clinical practice. [18] Additionally, in an Egyptian study claims that the variability in patient response emphasizes the need for individualized treatment approaches. Ongoing research and long-term studies are essential to fully understand the benefits and limitations of tafamidis therapy in diverse patient populations. [19]

Emerging Therapies

Recent developments in the treatment landscape for ATTR-CM include the FDA/EMA approvals of new therapies worldwide such as acoramidis (Attruby) by BridgeBio and positive trial results for vutrisiran by Alnylam Pharmaceuticals. These advancements indicate a growing array of therapeutic options for patients with ATTR-CM.[20]

Rationale

While several advancements have been made in diagnosing and treating ATTR-CM, gaps in knowledge still remain. [redacted]

The disease may present with variable presentations based on genetic and non-genetic forms (wild-type vs. hereditary). Understanding the mechanisms driving this heterogeneity remains still incomplete. While tools like bone scintigraphy and cardiac MRI have improved diagnostic options, there is limited standardization of non-invasive diagnostic criteria and biomarkers for early disease detection. ATTR-CM remains underdiagnosed due to its nonspecific symptoms that overlap with other forms of heart failure and cardiomyopathies. The lack of awareness among cardiologists often results in delayed or missed diagnoses. Therefore, our study aims to address these gaps by providing comprehensive real-world data on Turkish patients with ATTR-CM treated with tafamidis.

This noninterventional study is designated as a PASS and is conducted voluntarily by Pfizer.

7. RESEARCH QUESTION AND OBJECTIVES

The aim of this study is to describe the clinical characteristics and outcomes of patients with ATTR amyloid cardiomyopathy (ATTR-CM) treated with tafamidis at a single center in Turkey.

Primary Objective:

- To describe the rate of hospitalizations and emergency admissions due to chronic heart failure in the patients treated with Tafamidis

Secondary Objectives:

- To describe the rate of cardiovascular-related hospitalizations and overall survival among patients treated with Tafamidis during Tafamidis exposure time period.
- To describe the presence of cardiac and extracardiac red flags (Extra-cardiac Red Flags: Carpal Tunnel Syndrome, Peripheric Neuropathy, Lumbar Stenosis, Renal Insufficiency, Cardiac Red

Flags: Elevated NT Pro BNP and troponin, Pseudo infarct Sign, Voltage Mismatch, Conduction Disorder, Increased Wall Thickness (Interventricular septum), Pericardial Effusion)) in patients with ATTR - CM treated with tafamidis during the baseline period prior to initiation of tafamidis treatment.

- To describe the clinical and functional outcomes in ATTR CM patients during the baseline period prior to tafamidis treatment and during the follow-up period of treatment with tafamidis such as HRQoL measures, concomitant medication, biomarkers (NT pro-BNP, troponin), echocardiographic parameters (6 min walk test, Kansas City Cardiomyopathy Questionnaire Score, Creatinine eGFR, Ejection Fraction, Interventricular Septum, E/A ratio, E/e' mean ratio, Tricuspid Regurgitation Velocity, Tricuspid Annular Plane Systolic Excursion).

8. RESEARCH METHODS

8.1. Study Design

This is a single-center, retrospective, non-interventional cohort (Figure 1) study. Pseudonymized data will be entered in CRF from individual patient medical records maintained in a single center in Turkey according to clinical practice. Data will be collected using electronic case report form (eCRF). Subjects include diagnosed with ATTR-CM in the January 2023 – January 2025 and treated with tafamidis. For all patients, the 6-month period before the index date when tafamidis is started will be considered as the baseline period, and the relevant parameters will be collected from this period. From the start of tafamidis, quarterly follow up of the patients is defined as treatment period and clinical and functional outcomes, hospitalization and emergency admission due to CHF, CV related hospitalizations and mortality data will be obtained from this treatment period. Tafamidis treatment discontinuation is defined as interrupting the treatment for at least 10 days or completely stopping it, which will be determined by the primary investigator from the EHR records (Electronic Health Record). Hospitalization and emergency admission due to CHF (Chronic Heart Failure) and CV (Cardiovascular) related hospitalizations and other variables are described in Section 9.3.1. and Table 1.

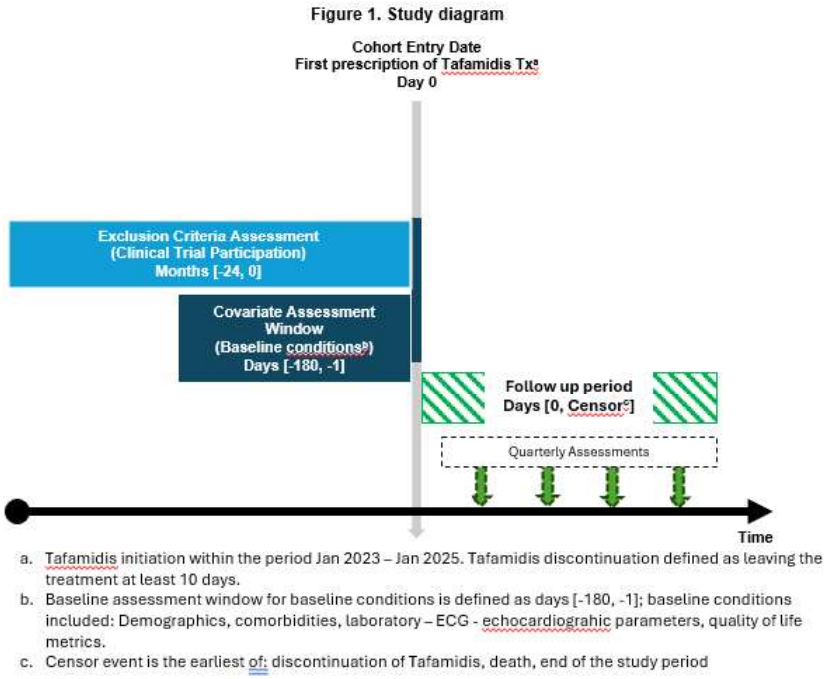

Figure 1. Study diagram

Rationale for the study design. The rationale for choosing a retrospective cohort design is that the data points of interest outlined in the study protocol are routinely collected in the Turkish clinical practice. Therefore, the retrospective approach will ensure that the dataset is available on the site. The single center participating in the study is highly specialized and skilled in the management of the target population.

This study is a cohort study because it involves following a group of ATTR CM patients who share common characteristics (in this case, the target population managed by the specialized center) over a period. Since the ATTR CM is a progressive disease, a cohort design will allow longitudinal evaluation of clinical outcomes and progression as managed in real world clinical practice. The cohort design was chosen instead of different study designs because it allows for the longitudinal evaluation of clinical outcomes and disease progression in patients with ATTR amyloid cardiomyopathy (ATTR-CM), which is essential for understanding the impact of tafamidis treatment over time. This design leverages routinely collected data in Turkish clinical practice, ensuring the availability of relevant information and reflecting real-world settings. Additionally, the specialized center's expertise in managing ATTR-CM patients enhances the reliability and applicability of the study results.

Strength of the study is described in the Section 9.7 (Strengths and Limitations of the Research Methods).

8.2. Setting

One Turkish Centre involved in the diagnosis and management of patients affected by ATTR-CM will participate in the study. This Centre is selected according to their clinical and research expertise, in

addition to the number of patients they are used to routinely caring for. A clinical research organization (CRO) on behalf of Pfizer will support the regulatory and administrative procedures to get authorization and the operations at Centre needed for data collection.

After regulatory authorization and local contract finalization, the study will be started at Centre. The site Study Team, including the Principal Investigator and additional identified personnel (e.g., research nurse, clinical study manager) will be trained on the protocol and planned study procedures, in addition to the use of the study-specific eCRF. Study data will be collected according to Section 9.1. Study Design and Figure 1 as patients are diagnosed with ATTR-CM between January 2023 and January 2025. For all patients, the 6-month period before the index date when tafamidis is started will be considered as the baseline period, and the relevant parameters will be collected from this period. Original data, routinely collected by means of specific tools (e.g., paper charts, electronic charts,) will be transferred to the eCRF by the Study Team in site.

Retrospective data from the patient's files/source data will be collected in the CRF up to the earliest date of: treatment discontinuation, death, lost to follow-up (the patient could not be observed and any more contacted at some time point during their follow-up) or end of the follow-up period of 30-Sep-2025 (inclusive). For the patients lost to follow-up, the end of their follow-up period will be the respective date of the last contact. In the case patients experience clinical events (e.g. hospitalization due to Chronic Heart Failure, safety event) starting before 30-Sep-2025 and continuing beyond end of the follow-up, the respective details until the end of the event should be included into the eCRF.

The CRO will ensure oversight and monitoring of the study at the site.

Approximately seventy (70) subjects who meet the inclusion and exclusion criteria detailed below will be included in the study. The inclusion and exclusion criteria were established based on the diagnosis of ATTR-CM disease and the evaluation of tafamidis treatment outcomes as outlined in the study objectives. The study population formed in light of these criteria is expected to represent the source population and the population of Turkey.

8.2.1. Inclusion Criteria

Patients must meet all the following inclusion criteria to be eligible for inclusion in the study:

1. ≥18 years old at the time of diagnosis of ATTR-CM,
2. Diagnosed with hereditary or wild type ATTR-CM confirmed non-invasively (Tc99m PYP bone scintigraphy and/or SPECT-CT.) in Samsun Education and Research Hospital – Cardiology Outpatient Clinic between 1 January 2023 – 1 January 2025,
3. Currently receiving or who have received tafamidis treatment for ATTR-CM,
4. Availability of complete medical records, including baseline clinical characteristics, treatment outcomes, and follow-up data from electronic health record systems of hospital or physical patient files.

8.2.2. Exclusion Criteria

Patients meeting any of the following criteria will not be included in the study:

1. Subjects with participation in studies involving investigational drugs within 24 months before the index dates.

8.3. Variables

Variables are described in the Table 1 below. If more than one measurement is taken during the baseline period, the most recent measurement to index date will be recorded to eCRF system.

Table 1. Variables of interests

Variable	Role	Assessment Period	Output	Data Source(s)	Operational Definition
Age at ATTR-CM diagnosis	Patient Baseline Characteristics, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records	Age will be calculated based on date of birth
Age at tafamidis start	Patient Baseline Characteristics, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records	Age will be calculated based on date of birth
Gender (M/F)	Patient Baseline Characteristics, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health h records or physical patient file	Male or female
Height (cm)	Patient Baseline Characteristics, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	Cm, (100-200)
Weight (kg)	Patient Baseline Characteristics, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	Kg, (30-150)
BMI (Body Mass Index)	Patient Baseline Characteristics, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	Categorized as underweight (<18.5) normal (18.0–24.9 kg/m^2), overweight (25.0–29.9 kg/m^2), or obesity (≥30 kg/m^2) (15-45)
Body Surface Area (automatic)	Patient Baseline Characteristics,	Baseline Assessment Period	Continuous	Electronic health records	Calculated with height and weight

	Secondary Objective			or physical patient file	
Hypertension (history of diagnosis:	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of hypertension (yes/no/missing) as documented in the source data
Diabetes Mellitus (history of diagnosis: Yes; or not: No)	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of diabetes mellitus (yes/no/missing) as documented in the source data
Hyperlipidemia	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of hyperlipidemia (yes/no/missing) as documented in the source data
Coronary Artery Disease	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of coronary artery disease (yes/no/missing) as documented in the source data
Heart Failure Stages	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Ordinal	Electronic health records or physical patient file	0-None 1-HFpEF (Heart Failure with preserved ejection fraction) 2-HFmrEF (Heart Failure with mildly reduced ejection fraction) 3-HFrEF (Heart Failure with reduced ejection fraction)
NYHA Class	Patient Baseline Comorbidities, Functional outcome, Secondary Objective	Baseline, Follow up Assessment Period	Ordinal	Electronic health records or physical patient file	A clinical assessment of a patient's cardiac disease and functional ability based on symptoms. Class 1-2-3-4

Chronic Obstructive Pulmonary Disease (COPD)	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of COPD (yes/no/missing) as documented in the source data
Atrial Fibrillation (AF)	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of atrial fibrillation (yes/no/missing) as documented in the source data
Aortic Stenosis	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Ordinal	Electronic health records or physical patient file	History of atrial fibrillation (yes/no/missing) as documented in the source data 0-None 1-Mild 2-Moderate 3-Severe
History of Surgical Aortic Valve Replacement (SAVR) or Transcatheter Aortic Valve Implantation (TAVI)	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of SAVR or TAVI (yes/no/missing) as documented in the source data
Carpal Tunnel Syndrome	Extracardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of Carpal Tunnel Syndrome (yes/no/missing) as documented in the source data
Peripheral Neuropathy	Extracardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of peripheral neuropathy (yes/no/missing) as documented in the source data
Lumbar Stenosis	Extracardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of lumbar stenosis (yes/no/missing)

					as documented in the source data
Pacemaker history	Patient Baseline Comorbidities, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	History of pacemaker implantation (yes/no/missing) as documented in the source data
Co-medications	Patient Baseline and Follow up Co-medications, Secondary Objective	Baseline, Follow up Assessment Period	Nominal	Electronic health records or physical patient file	Record of treatments used at the assessment timepoint (1: diuretics, 2: beta blockers 3: renin angiotensin axis inhibitors 4: anticoagulants 5: other)
Hemoglobin	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data g/dL, (5-20)
Creatinine	Extracardiac Red Flag, Functional Outcome, Secondary Objective	Baseline, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data mg/dL, (0.3-10)
eGFR (glomerular filtration rate)	Extracardiac Red Flag, Functional Outcome, Secondary Objective	Baseline, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	Automatic calculation with creatinine
Sodium	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data mmol/L, (100-200)
Potassium	Patient Baseline Laboratory	Baseline Assessment Period	Continuous	Electronic health records	As recorded in the source data mmol/L, (2-10)

	Parameters, Secondary Objective			or physical patient file	
ALT (Alanine transaminase) U/L	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data U/L (1-1000)
AST (Aspartate aminotransferase)	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data U/L, (5-1000)
Albumin	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data g/L, (10-200)
Modified BMI	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	(Albumin x BMI) (automatic calculation)
NT pro-BNP	Cardiac Red Flag, Functional Outcome, Secondary Objective	Baseline, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data ng/L, (100-100000)
Troponin	Cardiac Red Flag, Functional Outcome, Secondary Objective	Baseline, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data
Serum free light chain levels (Kappa) mg/L	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data mg/L, (5-1000)

Serum free light chain levels (Lambda) mg/L	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	As recorded in the source data mg/L, (5-1000)
Serum free light chain levels (Kappa/Lambda (automatic calculation)	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	free-kappa–lambda ratio (automatic calculation)
Serum immunofixation electrophoresis monoclonal gammopathy	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	As recorded in the source data yes/no
24h urine immunofixation monoclonal gammopathy	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	As recorded in the source data yes/no
Immunofixation/Immune electrophoresis monoclonal gammopathy	Patient Baseline Laboratory Parameters, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	As recorded in the source data yes/no
Heart Rate	Patient Baseline ECG Parameters, Secondary Objective	Baseline Assessment Period	Discrete	Electronic health records or physical patient file	(BPM) Beat per minute, resting, as recorded in the source data
Rhythm	Patient Baseline ECG Parameters, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Heart rhythm 1-Normal Sinus Rhythm 2-Atrial Fibrillation
Pseudo infarct Sign	Cardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	a Q-wave inversion on an electrocardiogram that mimics an

					acute myocardial infarction (present: Yes; or not: No)
Voltage Mismatch	Cardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Incompatibility between low QRS voltage on ECG and increased LV wall thickness on echocardiography (present: Yes; or not: No)
Conduction Disturbance	Cardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	The cardiac area refers to any abnormality in the electrical conduction system of the heart (present: Yes; or not: No)
EF = Ejection Fraction	Patient Baseline Echo Parameters, Functional Outcome, Secondary Objective	Baseline, Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	The percentage of blood that is ejected from the left ventricle of the heart with each contraction, as recorded in the source echocardiography data %
IVS (Interventricular Septum)	Patient Baseline Echo Parameters, Functional Outcome, Secondary Objective	Baseline, Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	The structure that separates the right and left ventricles of the heart, as recorded in the source echocardiography data mm
LVEDD (Left ventricle end diastolic diameter)	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The measurement of the diameter of the left ventricle at the end of diastole, as

					recorded in the source echocardiography data mm
LVESD (Left ventricle end systolic diameter)	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The measurement of the diameter of the left ventricle at the end of systole, as recorded in the source echocardiography data mm
PW (posterior wall)	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The thickness of the posterior wall of the left ventricle, measured during diastole, as recorded in the source echocardiography data mm
LAVI (Left atrial volume index)	Patient Baseline Echo Parameters, Functional Outcome, Secondary Objective	Baseline, Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	The volume of the left atrium normalized to body surface area, as recorded in the source echocardiography data mL/m^2
LVMI (Left ventricle mass index)	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The mass of the left ventricle normalized to body surface area, as recorded in the source echocardiography data mL/m^2
E/A Ratio (Early to atrial	Patient Baseline Echo	Baseline, Index,	Continuous	Electronic health records	The ratio of peak velocity blood

filling velocity ratio)	Parameters, Functional Outcome, Secondary Objective	Follow up Assessment Period		or physical patient file	flow from left ventricular relaxation in early diastole to peak velocity flow in late diastole caused by atrial contraction, as recorded in the source echocardiography data
e' Lateral (mitral annulus wave)	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The early diastolic velocity of the lateral mitral annulus, as recorded in the source echocardiography data mm/s
e' Septal (mitral annulus wave) mm/s	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The early diastolic velocity of the septal mitral annulus, as recorded in the source echocardiography data mm/s
E/e' Lateral Ratio (Early mitral inflow velocity and mitral annular early diastolic velocity ratio)	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The ratio of early mitral inflow velocity to the early diastolic velocity of the lateral mitral annulus, as recorded in the source echocardiography data
E/e' Septal Ratio (Early mitral inflow velocity and mitral annular	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	The ratio of early mitral inflow velocity to the early diastolic velocity of the

early diastolic velocity ratio)					septal mitral annulus, as recorded in the source echocardiography data
E/e' Mean Ratio (Early mitral inflow velocity and mitral annular early diastolic velocity ratio)	Patient Baseline Echo Parameters, Functional Outcome, Secondary Objective	Baseline, Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	The average ratio of early mitral inflow velocity to the early diastolic velocity of the mitral annulus, as recorded in the source echocardiography data
Tricuspid Regurgitation Velocity	Patient Baseline Echo Parameters, Functional Outcome, Secondary Objective	Baseline, Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	The peak velocity of blood flow through the tricuspid valve during regurgitation as recorded in the source echocardiography data m/s
Diastolic Dysfunction Stage	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Ordinal	Electronic health records or physical patient file	The impairment of the heart's ability to relax and fill during diastole. It is typically graded based on echocardiographic findings (stage 1-2-3)
5-5-5 Findings	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	All e', a', s' velocities < 5 cm/s. This is a clue to the diagnosis of Cardiac Amyloidosis (present: Yes; or not: No)

TAPSE (Tricuspid Annular Plane Systolic Excursion)	Patient Baseline Echo Parameters, Functional Outcome, Secondary Objective	Baseline, Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	The measurement of the displacement of the tricuspid annulus during systole, reflecting right ventricular function as recorded in the source echocardiography data mm
sPAP (Systolic Pulmonary Artery Pressure	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Continuous	Electronic health records or physical patient file	Measurement of the pressure in the pulmonary artery during systole, as recorded in the source echocardiography data mmHg
Pericardial Effusion	Cardiac Red Flag, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Accumulation of fluid in the pericardial sac exceeding the physiological amount (present: Yes; or not: No)
Biatrial Dilatation	Patient Baseline Echo Parameters, Secondary Objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Enlargement of both atria, (present: Yes; or not: No)
Valve Thickening	Cardiac Red Flag, Secondary objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Increased thickness of heart valves, assessed via echocardiography (present: Yes; or not: No)
Tc99m PYP uptake; Perugini score	Patient Baseline Clinical Parameters,	Baseline Assessment Period	Ordinal	Electronic health records or physical patient file	Semi-quantitative visual scoring system for Tc99m PYP uptake, used

(PYP cardiac Scintigraphy Grade)	Secondary objective				to differentiate cardiac ATTR from AL amyloidosis (Grade 0-1-2-3)
Identification of hereditary vs. wild-type ATTR-CM (Wild - Variant)	Patient Baseline Clinical Parameters, Secondary objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Wild and Variant type of ATTR CM, this variable is defined by the TTR mutation test result, as recorded in the source data
TTR mutation variant (V30M, V122l, T60A, Ala81Thr, Arg34Gly, Other)	Patient Baseline Clinical Parameters, Secondary objective	Baseline Assessment Period	Nominal	Electronic health records or physical patient file	Mutation Type: V30M, V122l, T60A, Ala81Thr, Arg34Gly and other
6 min walking test m	Functional outcome, Secondary objective	Index, Follow up Assessment Period	Continuous	Electronic health records or physical patient file	Tests results as recorded in the source data
KCCQ Score per treatment (Kansas City Cardiomyopathy Questionnaire)	Functional outcome, Secondary objective	Index, Follow up Assessment Period	Ordinal	Electronic health records or physical patient file	Assessing physical limitations, symptoms, self-efficacy, social interference, and quality of life in heart failure patients.
Hospitalization due to CHF (Chronic Heart Failure)	Hospitalization data, Primary objective	Follow up Assessment Period	Nominal	Electronic health records or physical patient file	Admission to a hospital specifically for the treatment of Chronic Heart Failure will be recorded from electronic healthcare records trough international classification system codes and assessment of

					primary investigator, ICD codes in the EHR and assessment of the investigator will be used to classify the reason of hospitalization event as CHF, the ICD codes are defined at Section **9.3.1.**
Admission to the emergency department due to CHF (present: Yes; or not: No)	Emergency admission data, Primary objective	Follow up Assessment Period	Nominal	Electronic health records or physical patient file	Emergency admission will be recorded from electronic healthcare records trough international classification system codes and assessment of primary investigator, ICD codes in the EHR and assessment of the investigator will be used to classify the reason of admission of the emergency event as CHF, the ICD codes are defined at **Section 8.3.1.**
Cardiovascular related hospitalizations (present: Yes; or not: No)	Hospitalization data, Secondary objective	Follow up Assessment Period	Nominal	Electronic health records or physical patient file	Cardiovascular related hospitalizations will be recorded from electronic healthcare records trough international classification system codes and

					assessment of primary investigator, ICD codes in the EHR and assessment of the investigator will be used to classify the reason of hospitalization event as cardiovascular related, the ICD codes are defined at **Section 8.3.1.**
All-cause Mortality (present: Yes, date of event; or not: No)	Mortality data, Secondary objective	Follow up Assessment Period	Nominal	Electronic health records or physical patient file	All-cause mortality including the date of the event (DD/MM/YYYY) will be recorded from electronic healthcare records
Tafamidis treatment prescription date	Primary and secondary objective	Index date	Continuous	Electronic health records or physical patient file	The date on which the patient receives the prescription of Tafamidis as recorded in the patient's medical record. The start of Tafamidis treatment is expected shortly after the date of prescription. (DD/MM/YYYY)
Tafamidis treatment end date	Primary and secondary objective	Follow up assessment period	Continuous	Electronic health records or physical patient file	The date on which a patient stops receiving Tafamidis treatment, recorded in the patient's medical records. (DD/MM/YYYY)

Any discontinuation of Tafamidis (Yes/No)	Primary and secondary objective	Follow up Assessment Period	Nominal	Electronic health records or physical patient file	The cessation of Tafamidis treatment for any reason (Yes/No)
Tafamidis discontinuation reason (1. Mortality, 2. Reimbursement rule, 3. Lost to follow up, 4. other)	Primary and secondary objective	Follow up Assessment Period	Ordinal	Electronic health records or physical patient file	Tafamidis Discontinuation Reason: (1. Mortality, 2. Reimbursement rule, 3. Lost to follow up, 4. other)

The dates of parameters obtained from tests such as ECG and echocardiography recorded at baseline, index, and follow-up dates will be entered into the eCRF system.

8.3.1. Definition of CHF-, CV- related hospitalizations and emergency admissions, and Red Flags

In our study, CHF and CV-related hospitalizations will be defined as following:

In this center and Turkish clinical practice, a **hospitalization or emergency admission due to chronic heart failure** is defined as an unplanned hospital admission due to worsening heart failure symptoms requiring intravenous diuretics, inotropic support, or other advanced heart failure therapies. In the electronic healthcare records international classification system codes (Version of ICD 10, such as I50 Heart Failure and its subcodes I50.1 Left ventricular failure, I50.2 Systolic Heart Failure, I50.3 Diastolic Heart Failure, I50.4 Combined Heart Failure, I50.8 Combined Heart Failure, etc.) for chronic heart failure hospitalization is defined and well recorded in this site. Emergency admissions for causes not related to chronic heart failure could not be accurately and completely captured; thus, these will not be recorded in CRF.

A **cardiovascular-related hospitalization** is defined as an unplanned hospital admission due to any cardiovascular event, in the electronic healthcare records international classification system codes such as I42 Cardiomyopathy, I44 Atrioventricular and left bundle branch block, I51 Complications and ill-defined descriptions of heart disease, I50 Heart Failure, etc.) . The primary investigator is going to evaluate all cardiovascular and chronic heart failure related hospitalizations in terms of general practice in Turkey.

We identified specific **cardiac** and **extra-cardiac** symptoms and signs indicative of cardiac amyloidosis, which are classified as **red flags**. These were organized based on the guidelines from the “Diagnosis and Treatment of Cardiac Amyloidosis: A Position Statement of the European Society of Cardiology” (Garcia-Pavia P, Rapezzi C, Adler Y, et al. Diagnosis and treatment of cardiac amyloidosis: A position statement of the ESC Working Group on Myocardial and Pericardial Diseases. *Eur Heart J* -2021;42(16):1554–1568.), some red flags mentioned in the position statement are not used or measured in daily clinical practice, therefore they are not planned to be included in the study.

Extra-cardiac Red Flags

- Carpal Tunnel Syndrome, Peripheric Neuropathy, Lumbar Stenosis, Renal Insufficiency (creatinine and GFR).

Cardiac Red Flags

- Elevated NT ProBNP and troponin, Pseudo infarct Sign, Voltage Mismatch, Conduction Disorder, Increased Wall Thickness (Interventricular septum), Pericardial Effusion.

8.4. Data Sources

This is a retrospective study of existing healthcare data collected; no participants will be actively enrolled and there will be no collection of any primary data. All pseudonymized data will be entered/ collected in a standardized case report form (CRF) designed specifically for this study. Data will be collected from individual patient medical records maintained in the Centre according to clinical practice in paper or electronic version. An electronic case report form (eCRF) will be utilized to ensure efficient data collection and management. Data collection will be performed by trained person who are familiar with the study protocol and the clinical data being collected. These individuals will be responsible for accurately entering the pseudonymized data into the eCRF.

The data collection process will involve reviewing patient medical records and extracting relevant information according to predefined criteria outlined in the study protocol. The pseudonymized data will be collected from site and will be processed for further analysis. For pseudonymization, each participant will have a specific code. Only the site investigators will be able to link the code with the participant.

Hospitalizations and emergency department visits due to CHF will be ascertained by the primary investigator using ICD codes and supporting hospital records. The primary investigator will verify that the event was primarily caused by chronic heart failure, based on the patients' history, laboratory results, and medications recorded in the discharge hospitalization or emergency summary form.

The validity of the recording and coding of the data will be ensured through several measures. First, the CRF will be designed to capture standardized clinical information, reducing variability in data entry. Second, the study site team responsible for data collection will undergo training to ensure consistency and accuracy in data abstraction. Third, quality review by CRO will be conducted to verify the accuracy and completeness of the recorded data.

8.5. Study Size

[redacted] Furthermore, to minimize selection bias, all patients diagnosed with ATTR-CM treated with tafamidis in this center and who meet the inclusion criteria will be eligible during the 24 months planned for enrollment procedures and until the target number of enrolled patients is reached according to the planned timeline.

8.6. Data Management

Data management will be performed by the CRO acting on behalf of Pfizer.

Data Collection and Entry: Patient data will be collected retrospectively from electronic health record systems maintained at the single participating center. The treatment protocol notes data will be collected in the eCRF. The data collection period is between January 2023 and September 2025. Patient data will be collected during the baseline diagnosis period and then throughout the treatment period (while the patient is under tafamidis treatment) during follow-ups period. The study does not involve any patient contact and will not impact the care patients receive. Preidentified data regarding the patients will be collected in electronic Case Report Forms eCRF and analyzed to accomplish the proposed study objectives.

Data will be collected on the e-CRF application, which will be prepared based on the CRF that will be developed specifically for this study. The e-CRF will meet the scientific, regulatory, and logistical requirements of the study before it is used to record data for this study. Before using the e-CRF, all users will receive training on the system and study specific training.

The single investigator from a single center and a site coordinator will be defined in the system. Form controls will be used for the appropriate data fields (for data fields with clear boundaries and content), this will prevent any data entry outside the limits set for the investigator. For data fields that cannot be limited by form controls but have boundaries, automatic form controls will be provided in the system.

All data required for the study, including nurses' measurements, patient demographics and necessary previous medical records will be recorded to the e-CRF manually by the investigator. The investigator is responsible for the accuracy of these manually recorded data.

During patient data entry, the faulty/missing data scan will be performed by the e-CRF automatically. In this way, any form with faulty or missing data will not be allowed to send data to e-CRF, and warning messages will be displayed on the computer screen to the person entering the data to ensure that he/she enter data correctly and completely.

Data cleaning and, if necessary, source data verification will be conducted. After cleaning, the database will be locked, and the data will be extracted in the appropriate format.

Patient identity should not be discernible from the data provided on the e-CRF and documents that will be stored as source data on patient file of this study. All data collected will be approved and signed by the investigator at the investigational center. This approval acknowledges the investigator's review and acceptance of the data as being complete and accurate.

Data Types: Collected data will include sociodemographic characteristics, clinical parameters, diagnostic results, treatment details, and patient outcomes, as detailed in the study protocol. The medical charts of all eligible patients were reviewed for the following data: demographic characteristics, lab tests, serum free light chain levels, Serum/urine protein/immunofixation electrophoresis, genetic tests, ECG, Echocardiography, Scintigraphy results SPECT results.

Data Entry: Data will be entered into a secure, password-protected electronic database ensuring compliance with relevant regulations and guidelines.

Data Validation: All data entries will be double-checked for accuracy by designated personnel. Any discrepancies will be discussed with investigator resolved by cross-referencing the source documents.

8.6.1. Case Report Forms/Data Collection Tools/Electronic Data Record

As used in this protocol, the term CRF should be understood to refer to either a paper form or an electronic data record or both, depending on the data collection method used in this study.

A CRF is required and should be completed for each included patient. The completed original CRFs are the sole property of Pfizer and should not be made available in any form to third parties, except for authorized representatives of Pfizer or appropriate regulatory authorities, without written permission from Pfizer. The investigator shall ensure that the CRFs are securely stored at the study site in encrypted electronic form and will be password protected to prevent access by unauthorized third parties.

The investigator has ultimate responsibility for the collection and reporting of all clinical, safety, and laboratory data entered on the CRFs and any other data collection forms (source documents) and ensuring that they are accurate, authentic/original, attributable, complete, consistent, legible, timely (contemporaneous), enduring, and available when required. The CRFs must be signed by the investigator or by an authorized staff member to attest that the data contained on the CRFs are true. Any corrections to entries made in the CRFs or source documents must be dated, initialed, and explained (if necessary) and should not obscure the original entry.

The source documents are the hospital or the physician's chart. In these cases, data collected on the CRFs must match those charts.

8.6.2. Record Retention

To enable evaluations and/or inspections/audits from regulatory authorities or Pfizer, the investigator agrees to keep records, including the identity of all participating patients (sufficient information to link records, eg, CRFs and hospital records), copies of all e-CRFs, safety reporting forms, source documents, detailed records of treatment disposition, and adequate documentation of relevant correspondence (eg, letters, meeting minutes, and telephone call reports). The records should be retained by the investigator according to local regulations or as specified in the clinical study agreement (CSA), whichever is longer. The investigator must ensure that the records continue to be stored securely for so long as they are retained.

If the investigator becomes unable for any reason to continue to retain study records for the required period (eg, retirement, relocation), Pfizer should be prospectively notified. The study records must be transferred to a designee acceptable to Pfizer, such as another investigator, another institution, or to an independent third party arranged by Pfizer.

Study records must be kept for a minimum of 15 years after completion or discontinuation of the study, unless or as required by applicable local regulations.

The investigator must obtain Pfizer's written permission before disposing of any records, even if retention requirements have been met.

8.7. Data Analysis

The anonymously collected data of each patient cohort will be analyzed with an exploratory data analysis approach. The de-identified dataset will be analyzed using statistical software. Descriptive

statistics, survival analyses, and exploratory methods will be employed to address study objectives. Outliers and data inconsistencies will be reviewed and addressed before final analysis. Only descriptive statistics will be reported and no effort to determine statistical significance will be made.

Hospitalization and emergency department admission rates attributable to chronic heart failure, as well as hospitalization rates for cardiovascular-related illness, will be expressed as events per patient-year to account for variable follow-up durations.

Overall survival will be described using the Kaplan–Meier method, appropriately accounting for differing follow-up times and censoring; competing risks will not be considered.

Cardiac and extra-cardiac "red flags," together with clinical and functional outcomes, will be described using means (standard deviations) for continuous variables and proportions for categorical variables.

Missing data will not be imputed; all analyses will be based on available data, with the number of missing observations reported for each variable where applicable.

Detailed methodology for summary and statistical analyses of data collected in this study will be documented in a statistical analysis plan (SAP), which will be dated, filed, and maintained by the sponsor. The SAP may modify the plans outlined in the protocol; any major modifications of primary endpoint definitions or their analyses would be reflected in a protocol amendment.

8.8. Quality Control

Before the study launch, study personnel will review data collection and processing procedures, with all study-related trainings recorded in a training log.

The principal investigator and a designated CRO team will monitor the study data for accuracy and completeness, conducting periodic reviews to ensure adherence to the study protocol, quality of data collection, and completeness of datasets, following SOPs for all aspects of data collection, entry and data analysis consistency and reliability. The analytic dataset consisting of the collected data will be validated by the principal Investigator or designee who will verify the following:

- Review of illogical data or atypical values (incl. the values out of range and with different units/ information of what is indicated in Table 1. Variables of interests);
- Verification of conflicting data;
- Review of very high percentages of response as "not known" or "data not available";
- Verification of accuracy of the collected data by comparison with the source documents.

CRO is responsible for following their own SOPs as well as Pfizer's SOPs to ensure data quality and integrity, including archiving of statistical programs, appropriate documentation of data cleaning and validation of derived variables. Guidelines for data collection and data quality checks will be described in the Monitoring and Data Quality Oversight Plan.

A record of data quality problems and resolutions will be kept through documented meeting minutes.

All inconsistencies and/or data quality issues will be documented. Revisions of study documents will be noted in order to capture the change made, the change date, identification of the individual making

the change, as well as noting any further actions to be taken to identify and/or resolve additional data quality problems of this type.

Electronic Data will be stored in a secure, password-protected database with restricted access in compliance with all local applicable laws and regulations, with regular backups to prevent loss, physical paper version records will be stored in a locked, secure location accessible only to authorized personnel. Statistical programming and datasets used to generate study results will be archived with version control, ensuring reproducibility of analyses.

Data collection and processing will be limited to necessary data for study objectives, with data pseudonymized to protect participant privacy.

Statistical analysis scripts and programming used for the study will be independently reviewed by a biostatistician for accuracy and compliance with study objectives. All statistical code and datasets will be archived and available for audit as required.

8.9. Strengths and Limitations of the Research Methods

This study will provide insights into the clinical outcomes of ATTR-CM patients treated with tafamidis in Turkey. This localized focus on a unique population adds to the limited data on regional variations in the disease and treatment outcomes.

This study will have several limitations. Being a single-center study, the findings may not be generalizable to other centers, regions, or healthcare systems with different patient populations and practices. Medical records may have missing or incomplete information (e.g., certain biomarkers, imaging results, or medication adherence data).

Ascertainment of CHF hospitalization will be done by a principle investigator using all available records; potential misclassification of the reasons for hospitalizations is possible. To partially address the issue of misclassification, all CV hospitalizations will be captured and recorded in CRF as well.

Emergency admissions for causes other than CHF will not be captured and analyzed. There is a potential, that some of the emergency admissions could be related to cardio-vascular causes, but full verification may not be possible; this will remain a limitation of the current study.

Only patients diagnosed and treated at the single participating center will be included, which could lead to a selection bias. Patients lost to follow-up or those who did not complete tafamidis treatment may not be fully represented.

The design of the study does not include a comparative arm and does not allow for robust causal conclusions related to tafamidis treatment effectiveness or safety.

8.10. Other Aspects

Not Applicable

9. PROTECTION OF HUMAN PARTICIPANTS

9.1. Patient Information

All parties will comply with all applicable laws, including laws regarding the implementation of organizational and technical measures to ensure protection of patient personal data. Such measures will include omitting patient names or other directly identifiable data in any reports, publications, or other disclosures, except where required by applicable laws.

The personal data will be stored at the study site in encrypted electronic form and/or paper version and will be password protected or secured in a locked room to ensure that only authorized study staff have access. The study site will implement appropriate technical and organizational measures to ensure that the personal data can be recovered in the event of disaster. In the event of a potential personal data breach, the study site shall be responsible for determining whether a personal data breach has in fact occurred and, if so, providing breach notifications as required by law.

To protect the rights and freedoms of natural persons with regard to the processing of personal data, when study data are compiled for transfer to Pfizer and other authorized parties, patient names will be removed and will be replaced by a single, specific, numerical code, based on a numbering system defined by Pfizer. All other identifiable data transferred to Pfizer or other authorized parties will be identified by this single, patient-specific code. The investigator site will maintain a confidential list of patients who participated in the study, linking each patient's numerical code to his or her actual identity. In case of data transfer, Pfizer will maintain high standards of confidentiality and protection of patients' personal data consistent with the clinical study agreement and applicable privacy laws.

9.2. Patient Consent

As this study does not involve data subject to privacy laws according to applicable legal requirements, obtaining informed consent from patients by Pfizer is not required.

9.3. Institutional Review Board (IRB)/ Ethics Committee (EC)

There must be prospective approval of the study protocol, protocol amendments, and other relevant documents (e.g., informed consent forms if applicable) from the relevant IRBs/ECs. All correspondence with the IRB/EC must be retained. Copies of IRB/EC approvals must be forwarded to Pfizer.

9.4. Ethical Conduct of the Study

The study will be conducted in accordance with legal and regulatory requirements, as well as with scientific purpose, value, and rigor and follow generally accepted research practices described in:

- Guidelines for Good Pharmacoepidemiology Practices (GPP). Public Policy Committee, International Society of Pharmacoepidemiology. Pharmacoepidemiology and Drug Safety 2016; 25:2-10.

- Good practices for real-world data studies of treatment and/or comparative effectiveness: Recommendations from the joint ISPOR-ISPE Special Task Force on real-world evidence in health care decision making
- International Ethical Guidelines for Epidemiological Studies issued by the Council for International Organizations of Medical Sciences (CIOMS)
- The ENCePP Code of Conduct for scientific independence and transparency in the conduct of pharmacoepidemiological and pharmacovigilance studies
- European Medicines Agency (EMA) European Network of Centres for Pharmacoepidemiology and Pharmacovigilance (ENCePP) Guide on Methodological Standards in Pharmacoepidemiology http://www.encepp.eu/standards_and_guidances/methodologicalGuide.shtml

10. MANAGEMENT AND REPORTING OF ADVERSE EVENTS/ADVERSE REACTIONS REQUIREMENTS

This study protocol requires human review of patient-level unstructured data; unstructured data refer to verbatim medical data, including text-based descriptions and visual depictions of medical information, such as medical records, images of physician notes, neurological scans, x-rays, or narrative fields in a database. The reviewer is obligated to report safety events (AEs/SAEs) with explicit attribution to any Pfizer product that appear in the reviewed information (defined per the patient population and study period specified in the protocol).

Explicit attribution is not inferred by a temporal relationship between drug administration and an AE but must be based on a definite statement of causality by a healthcare provider linking drug administration to the AE with such causality documented in the medical chart.

The requirements for reporting safety events, as defined below, on the "*Non-Interventional Study Adverse Event Report Form for Protocols with Stipulated Active Collection of Adverse Events*", herein after referred to as the NIS AEM Report Form are as follows:

- All safety events with explicit attribution to **any Pfizer drug** that appear in the reviewed information must be recorded on the CRF and reported, within 24 hours of awareness, to Pfizer Safety using the NIS AEM Report Form.
- Scenarios involving drug exposure, including exposure during pregnancy[(a)], breastfeeding, medication error, overdose, misuse, extravasation, lack of efficacy, and occupational exposure, and off-label use associated with the use of any Pfizer product must be reported, within 24 hours of awareness, to Pfizer Safety using the NIS AEM Report Form.

  [(a)] Exposure during pregnancy (EDP) reports are reportable using the NIS AEM Report Form and the EDP Supplemental Form, irrespective of the presence of an associated safety event.

  For exposure during pregnancy in studies exclusively of pregnant people, data on the exposure to tafamidis during pregnancy, are not reportable. However, if the mother or the fetus experiences any safety event (either serious or non-serious), the event must be reported without the event EDP.

For these safety events with an explicit attribution or scenarios involving exposure to any Pfizer product, the safety information identified in the unstructured data reviewed is captured in the Event Narrative section of the report form, and constitutes all clinical information known regarding them. No follow-up will be conducted.

All the demographic fields on the NIS AEM Report Form may not necessarily be completed, as the form designates, since not all elements will be available due to privacy concerns with the use of secondary data sources. While not all demographic fields will be completed, at the very least, one patient identifier (e.g., gender, age as captured in the narrative field of the form) will be reported on the NIS AEM Report Form, thus allowing the report to be considered a valid one in accordance with pharmacovigilance legislation. All identifiers will be limited to generalities, such as the statement "A 35-year-old female..." or "An elderly male...". Other identifiers will have been removed.

Additionally, the onset/start dates and stop dates for "Illness," "Study Drug," and "Drug Name" may be documented in month/year (mmm/yyyy) format rather than day/month/year (DD/MMM/YYYY) format."

All site/research staff members must complete the following Pfizer training requirements:

- *"Your Reporting Responsibilities (YRR) with Supplemental Topics."*

This training must be completed by research staff members prior to the start of data collection. All trainings include a "Confirmation of Training Statement" (for signature by the trainee) as a record of completion of the training, which must be kept in a retrievable format. Copies of all signed training statements must be provided to Pfizer.

11. PLANS FOR DISSEMINATING AND COMMUNICATING STUDY RESULTS

The results of the study are intended to be published in the scientific literature or presented at a scientific congress. The study team will follow the International Committee of Medical Journal Editors (ICMJE) criteria to determine authors, and all authors who meet these criteria will be offered authorship.

In the event of any prohibition or restriction imposed (e.g., clinical hold) by an applicable competent authority in any area of the world, or if the investigator is aware of any new information which might influence the evaluation of the benefits and risks of a Pfizer product, Pfizer should be informed immediately.

12. REFERENCES

1. Hazenberg et al., Amyloidosis: a clinical overview, Rheumatic Disease Clinics of North America Volume 39, Issue 2, May 2013, Pages 323-345
2. O. Siddiqi, F. Ruberg Cardiac amyloidosis: An update on pathophysiology, diagnosis, and treatment., Trends in Cardiovascular Medicine, Volume 28, Issue 1, January 2018, Pages 10-21
3. Nijst&Tang, Recent advances in the diagnosis and management of amyloid cardiomyopathy. Fac Rev. 2021 Mar 24:10:31. doi: 10.12703/r/10-31. eCollection 2021.

4. Maurer MS, et al. Tafamidis treatment for patients with TTR amyloid cardiomyopathy. New England Journal of Medicine. 2018;379(11):1007-1016.
5. Asuman Argon et al., Cardiac Amyloidosis: Clinical Features, Pathogenesis, Diagnosis, and Treatment Turk Patoloji Derg 2024, 40:1-9
6. Connors LH, Sam F, Skinner M, Salinaro F, Sun F, Ruberg FL, Berk JL, Seldin DC. Heart Failure Resulting From Age-Related Cardiac Amyloid Disease Associated With Wild-Type Transthyretin: A Prospective, Observational Cohort Study. Circulation. 2016 Jan 19;133(3):282-90. doi: 10.1161/CIRCULATIONAHA.115.018852
7. Ruberg FL, Grogan M, Hanna M, Kelly JW, Maurer MS. Transthyretin Amyloid Cardiomyopathy: JACC State-of-the-Art Review. J Am Coll Cardiol. 2019 Jun 11;73(22):2872-2891. doi: 10.1016/j.jacc.2019.04.003. PMID: 31171094; PMCID: PMC6724183.
8. Maurer et al, Genotype and Phenotype of Transthyretin Cardiac Amyloidosis: THAOS (Transthyretin Amyloid Outcome Survey) JACC, Volume 68, Issue 2, Pages 161–172; 2016
9. Connors LH, et al. T60A variant TTR amyloidosis. Amyloid. 2003;10(3):160-163.
10. González-López E, et al. Wild-type TTR amyloidosis as a cause of heart failure with preserved ejection fraction. European Heart Journal. 2015;36(44):2585-2594.
11. Omar F. AbouEzzeddine et al. Prevalence of Transthyretin Amyloid Cardiomyopathy in Heart Failure with Preserved Ejection Fraction, JAMA Cardiol. 2021;6(11):1267-1274. doi:10.1001/jamacardio.2021.3070
12. Grogan M, et al. TTR cardiac amyloidosis: natural history and treatment outcomes. Journal of the American College of Cardiology. 2016;68(10):1014-1020.
13. Garcia-Pavia P, Rapezzi C, Adler Y, et al. Diagnosis and treatment of cardiac amyloidosis: a position statement of the ESC Working Group on Myocardial and Pericardial Diseases. *Eur Heart J*. 2021;42(16):1554-1568. doi:10.1093/eurheartj/ehab072
14. Gillmore JD, Maurer MS, Falk RH, et al. Nonbiopsy Diagnosis of Cardiac Transthyretin Amyloidosis. *Circulation*. 2016;133(24):2404-2412. doi:10.1161/CIRCULATIONAHA.116.021612Miller RJH, Cadet S, Mah D, Pournazari P, Chan D, Fine NM, Berman DS, Slomka PJ. Diagnostic and prognostic value of Technetium-99m pyrophosphate uptake quantitation for transthyretin cardiac amyloidosis. J Nucl Cardiol. 2021 Oct;28(5):1835-1845. doi: 10.1007/s12350-021-02563-4. Epub 2021 Mar 10. PMID: 33689152; PMCID: PMC8497047.
15. Kristen AV, et al. Cardiac Amyloid Load: A Prognostic and Predictive Biomarker in Patients with Light-Chain Amyloidosis. J Am Coll Cardiol. 2014;64(10):1022-1032.
16. Garcia-Pavia P, Kristen AV, Drachman B, et al. Survival in a Real-World Cohort of Patients With Transthyretin Amyloid Cardiomyopathy Treated With Tafamidis: An Analysis From the Transthyretin Amyloidosis Outcomes Survey (THAOS). *J Card Fail*. 2025;31(3):525-533. doi:10.1016/j.cardfail.2024.06.003
17. Ghoneem A, Bhatti AW, Khadke S, et al. Real-World Efficacy of Tafamidis in Patients With Transthyretin Amyloidosis and Heart Failure. *Curr Probl Cardiol*. 2023;48(6):101667. doi:10.1016/j.cpcardiol.2023.101667

18. Hussain K, Macrinici V, Wathen L, et al. Impact of Tafamidis on Survival in a Real-World Community-Based Cohort. *Curr Probl Cardiol*. 2022;47(12):101358. doi:10.1016/j.cpcardiol.2022.101358

19. Ogieuhi IJ, Ugiomoh OM, Muzofa K, Callender K, Ayodeji JD, Nnekachi NP, Thiyagarajan B, Uduigwome EO, Kapoor A, Odoeke MC, Mohamed RGH, Idahor C. Tafamidis therapy in transthyretin amyloid cardiomyopathy: a narrative review from clinical trials and real-world evidence. Egypt Heart J. 2024 Jul 10;76(1):90. doi: 10.1186/s43044-024-00517-y. PMID: 38985360; PMCID: PMC11236832.

20. Griffin JM, Grodin JL, Ruberg FL, Masri A, Hanna M, Maurer MS. Current Landscape of Therapies for Transthyretin Amyloid Cardiomyopathy. *JACC Heart Fail*. 2025;13(5):685-694. doi:10.1016/j.jchf.2025.03.017

13. LIST OF TABLES

14. LIST OF FIGURES

ANNEX 1. LIST OF STANDALONE DOCUMENTS

None

ANNEX 2. ENCEPP CHECKLIST FOR STUDY PROTOCOLS

Not required

ANNEX 3. ADDITIONAL INFORMATION

Not applicable

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