

## **Abstract**

Transthyretin (TTR) is a tetrameric protein synthesized primarily by the liver that acts as a physiological transport protein for retinol and thyroxine. TTR can misfold into pathogenic ATTR amyloid fibrils that deposit in the heart and nerves, causing a life-threatening cardiomyopathy (CM) and a progressive and debilitating polyneuropathy (PN). Recent therapeutic advances have resulted in the development of drugs that reduce TTR production. Patisiran was the first gene silencing medication licensed for the treatment of ATTR amyloidosis, and is a small interfering RNA that disrupts the complementary mRNA and inhibits TTR synthesis. Encouraging results following use of patisiran for the treatment of patients with ATTR-PN, resulted in increasing interest in the use of patisiran for the treatment of ATTR-CM. Various studies have demonstrated improvements across a wide range of cardiac biomarkers following treatment with patisiran and have changed the perception of ATTR-CM from previously being thought of as a terminal disease process, to now being regarded as a treatable disease. These successes represent a huge milestone and have the potential to revolutionise the landscape of treatment for ATTR-CM. However, the long-term safety of patisiran and how best to monitor cardiac response to treatment remain to be determined.

**Key words:** Transthyretin amyloidosis, ATTR cardiomyopathy, Patisiran,

## Introduction

The systemic amyloidoses are a group of heterogeneous diseases characterised by the deposition of insoluble amyloid fibrils within the extracellular matrix of multiple organs throughout the body. Amyloid fibrils are formed from the misfolding of amyloid precursor proteins (APP) into insoluble, beta-pleated sheets, resistant to proteolysis; they are characterised histologically by apple-green birefringence when stained with Congo Red and examined under cross-polarised light.<sup>1,2</sup> Transthyretin amyloidosis, better known as ATTR amyloidosis, occurs secondary to the misfolding of the transthyretin (TTR) protein. TTR is synthesised by the liver, and circulates as a tetramer acting as a physiological vehicle for the transportation of retinol and thyroxine. ATTR amyloid fibrillogenesis occurs when the stable tetrameric form of TTR becomes unstable and dissociates into monomers or is cleaved into fragments, the substrate from which the fibrils are derived; disease occurs when the aggregation of amyloid fibrils disrupts the structure and function of an affected organ.<sup>3,4</sup> The factors that determine organ tropism of amyloid deposits remain uncertain. However, electron microscopic examination of cardiac and nerve biopsies has demonstrated basement membrane thickening or reduplication which is associated with formation of advanced glycosylation end products, thought to potentially play a role in promoting amyloid formation and fibril accumulation.<sup>5</sup>

The formation of ATTR amyloid fibrils either occurs secondary to a pathological process that is associated with age-related homeostatic mechanisms, and is known as wild-type ATTR (ATTRwt), or occurs secondary to an inherited single point mutation in the *TTR*-gene, when it is known as hereditary ATTR or variant ATTR (ATTRv) amyloid.<sup>6</sup> The clinical characteristics depend on the underlying inherited genetic mutation. ATTRwt amyloidosis predominantly affects males and is typically seen in patients over 65 years of age, although it may present as young as age 50 years.<sup>7</sup>

Myocardial deposition of ATTR fibrils causes an infiltrative and restrictive cardiomyopathy, typified by increased biventricular wall thickness and reduced myocardial compliance. Initially, stiffening of the myocardium results in increased left ventricular filling pressures and diastolic dysfunction. Evolution of the disease process leads to systolic impairment, which begins with longitudinal systolic dysfunction and is followed by radial systolic dysfunction. Amyloid infiltration begins in basal segments of the left ventricle and often results in a characteristic “bull’s eye” strain pattern on echocardiography, with an increased septal apical-to-basal strain ratio and relative apical sparing (Figure 1). Cardiac amyloid infiltration also results in characteristic features on cardiac magnetic resonance (CMR) imaging, with an elevated myocardial native-T1, biventricular circumferential subendocardial/transmural late gadolinium enhancement and an elevated myocardial extracellular volume (ECV) (Figure 2).<sup>8-10</sup> Cardiac manifestations of amyloid infiltration also include aortic stenosis, conduction disease and atrial fibrillation with an increased risk of thrombus formation.<sup>3,11</sup> Extra-cardiac sites of amyloid infiltration can result in carpal tunnel syndrome, lumbar spine stenosis and tendinopathies.<sup>12-14</sup> ATTRv amyloidosis may present at a younger age with a mixed clinical phenotype comprising ATTR cardiomyopathy (ATTR-CM) and a peripheral polyneuropathy (ATTR-PN). Patients presenting with polyneuropathy typically develop a progressive and debilitating, length dependant, axonal sensory-motor peripheral polyneuropathy and/or autonomic neuropathy. The multisystem nature of ATTR amyloidosis is often best managed with multidisciplinary input from cardiologists, neurologists and orthopaedists.<sup>15</sup> However, despite polyneuropathy causing severe debilitating symptoms, cardiac amyloid infiltration remains the main cause of mortality (Table 1).<sup>16,17</sup>

Traditionally, ATTR amyloidosis was considered a rare disease, but developments in diagnostics and increased awareness of the condition have resulted in a substantial increase in diagnoses over the last two decades. Increased recognition of ATTR amyloidosis has been

driven in part by a non-biopsy diagnostic algorithm that utilises the impressive sensitivity of bone scintigraphy for identification of cardiac ATTR amyloid deposits. Characteristic features of cardiac amyloidosis on echocardiography or CMR, combined with grade 2 or 3 cardiac uptake on bone scintigraphy, in the absence of biochemical evidence a monoclonal gammopathy is diagnostic of ATTR-CM.<sup>18</sup> This diagnostic algorithm has since been validated in several studies, including a large multicentre study that confirmed a high specificity and positive predictive value.<sup>19</sup> In cases in which the non-biopsy criteria are not fulfilled, diagnosis is confirmed with either grade 2 or 3 cardiac uptake on bone scintigraphy and ATTR amyloid in an extra-cardiac biopsy, or direct endomyocardial biopsy proof of ATTR-amyloid (Table1). The National Amyloidosis Centre (NAC) is the single centre in the United Kingdom commissioned for the diagnosis and monitoring of amyloidosis. Data from this centre has demonstrated a dramatic increase in the number of ATTR-CM diagnoses over the last 20 years, from 35 cases between 2002-2006, to 968 cases between 2017-2021.<sup>20</sup> A similar observation has been documented in Sweden, whereby the prevalence of ATTR-CM has increased from 1/100,000 to 5/100,000 over a 10 year period.<sup>21</sup> However, the true prevalence of ATTR-CM remains unknown, and with post mortem examinations revealing ATTR amyloid deposits in one quarter of cardiac histological samples in elderly patients,<sup>22</sup> and large population studies demonstrating certain *TTR*-gene variants to be endemic in specific populations, the true prevalence is likely to be significantly higher than that reported in the medical literature.<sup>23</sup>

Before the availability of specific disease-modifying medicines, patients with p.(Val50Met) ATTRv-PN were successfully treated through liver transplantation. Following orthotopic liver transplantation variant TTR is rapidly cleared from the systemic circulation, due to its short plasma half-life, and replaced by wild-type TTR since there is no extrahepatic source of circulating TTR. Halting hepatic variant TTR production stops ATTRv fibril formation and subsequent accumulation within the nervous system, and thus slows or prevents further

deterioration of neuropathic symptoms. Successes following liver transplantation supported the premise that reducing amyloidogenic TTR production could stabilise the disease process and prevent further deterioration.<sup>24</sup>

The last decade has seen the development of multiple pharmacological therapeutic agents that can be utilised in the treatment of ATTR amyloidosis. Initially, these agents were designed to induce targeted stabilisation of the TTR tetramer to prevent it from dissociating into amyloidogenic monomers or being cleaved into amyloidogenic fragments. This resulted in the development of tafamids, a small molecule TTR stabiliser, which was shown to reduce all-cause mortality in the seminal ATTR-ACT trial, and subsequently licensed as the only available treatment of ATTR-CM.<sup>25</sup> More recently, medicines have been developed with the aim of suppressing TTR production by specifically targeting and suppressing hepatic TTR synthesis. It is noteworthy that in all types of systemic amyloidosis in which reduction of the circulating APP (e.g., serum amyloid A protein in AA amyloidosis and monoclonal immunoglobulin light chain protein in AL amyloidosis) is achievable, it has been associated with improved patient outcomes. Indeed, there is a strong correlation between the magnitude of 'knockdown' of the circulating APP and clinical outcomes.<sup>26-28</sup> Studies with serial SAP scintigraphy,<sup>29</sup> a quantitative nuclear medicine technique which visualises visceral amyloid deposits, and with serial ECV measurement by CMR<sup>30</sup> have shown that substantial reduction of circulating APP concentration often leads to gradual amyloid regression from both the viscera and the heart. It is thus anticipated that deep reductions in circulating TTR will be associated with regression of amyloid and better outcomes in patients with ATTR amyloidosis. This treatment paradigm led to the development of patisiran, a first generation small interfering RNA (siRNA) that targets and degrades the mRNA that codes for the TTR protein.<sup>31,32</sup> This review will explore in detail the use of patisiran for the treatment of ATTR-CM, and also provide insights in future perspectives.

## **Patisiran**

### ***Mechanism of action and pharmacokinetics***

Patisiran was the original siRNA developed specifically to treat patients with ATTR amyloidosis. siRNAs comprise double-stranded RNA molecules that bind complementary mRNA molecules and cause degradation, hence interfering with protein synthesis. Following administration, siRNAs must be transported into the intracellular cytoplasm of the intended cells, while avoiding breakdown from phagocytes and nucleases in the circulation and also avoiding excretion.<sup>33-35</sup> Patisiran is formulated as an encapsulated lipid nanoparticle that protects the siRNA molecule. The lipid nanoparticle is coated in apolipoprotein E, which binds to the low-density lipoprotein on the hepatocyte membrane to ensure targeted delivery. Once released into the hepatocyte intracellular cytoplasm, the double-stranded siRNA molecule divides into two single-stranded RNA molecules that bind to a genetically preserved sequence in the 3' untranslated portion of the complementary wild-type or variant *TTR* mRNA. The binding process triggers Argonaute slicer protein activation, which subsequently degrades the mRNA, thereby inhibiting TTR synthesis.<sup>31,32</sup>

Patisiran is administered as an intravenous infusion of 0.3mg/kg every 3 weeks, with single administration achieving a maximum reduction in serum TTR concentration of 88%, and a steady plasma level being achieved by 24 weeks.<sup>36</sup> Patisiran is metabolised into nucleotides and has an elimination half-life of 3±2 days, with less than 1% being excreted unchanged in the urine. Mild or moderate renal impairment (chronic kidney disease stage 1-3) and mild hepatic impairment do not affect the pharmacokinetics of patisiran, or its ability to induce TTR knockdown, and patisiran does not interfere with cytochrome P450 activity (Table 2).<sup>31</sup>

### ***Clinical trials***

Following a successful phase I clinical trial,<sup>36</sup> a phase II clinical trial of 29 patients with ATTRv-PN demonstrated that after 2 doses of patisiran the mean reduction in serum TTR from baseline was 85% and the maximum reduction was 96%.<sup>37</sup> In a 24 month open label extension of 27 patients, 11 of whom had concomitant cardiac involvement; patisiran treatment halted progression of neurological disease and stabilised cardiac disease, with there being no significant changes in serum cardiac enzymes or echocardiographic parameters.<sup>38</sup>

The APOLLO phase III trial was a double-blind, placebo-controlled, randomised study of 225 patients with ATTRv-PN, and was designed to evaluate the safety and efficacy of patisiran. After 18 months of treatment, there was a significant improvement from baseline in the modified Neuropathy Impairment Score +7 (mNIS+7), Norfolk Quality of Life Questionnaire-Diabetic Neuropathy (Norfolk QOL-DN) score and gait speed in the group treated with patisiran compared to the placebo group.<sup>28</sup> Patients treated with patisiran demonstrated a significant improvement across all components of the COMPASS-31 scale, (which is designed to assess the burden of autonomic neuropathy) compared to those on placebo.<sup>39</sup> Following the success of the APOLLO trial, in 2018 patisiran was approved for the treatment of ATTRv-PN by both the European Commission (EC) and the United States Food and Drug Administration (FDA).<sup>31,32</sup>

### ***Cardiac outcomes***

A post-hoc analysis was carried out on a sub-set of 126 patients enrolled in the APOLLO trial considered to have cardiac involvement based on prespecified criteria of a left ventricular (LV) wall thickness  $\geq 13$ mm in the absence of hypertension or significant aortic valve disease, to assess the efficacy of patisiran in patients with ATTR-CM accompanying ATTR-PN (i.e., ATTR-Mixed). Treatment with patisiran was associated with a reduced mean left ventricular wall thickness and relative wall thickness at 18 months compared with placebo. Patisiran

treatment also resulted in an increased left ventricular end diastolic volume, improved global longitudinal strain and cardiac output. The patisiran group had a significant reduction in serum N-terminal pro B-type natriuretic peptide (NT-proBNP) and significant increase in 10-meter walk test gait speed when compared to patients in the placebo group. The favourable changes in both cardiac structure and function were associated with a lower combined event rate of all-cause hospitalisation and mortality in the treatment group compared to placebo. However, this post-hoc analysis does have significant limitations. The prespecified definition of cardiac involvement does not adhere with published diagnostic recommendations, and despite being sensitive, lacks specificity. A relatively small cohort of patients were included, with only 36 patients in the placebo arm, and due to the post-hoc design, the analysis was unblinded. Lastly, despite a significantly lower all-cause hospitalisation and mortality rate in the treatment group, there was no significant difference in cardiovascular hospitalisations and mortality, which raises the possibility that observed differences could be due to treatment of the underlying polyneuropathy rather than cardiomyopathy. Nevertheless, this analysis was the first to suggest that patisiran may halt the progression of cardiomyopathy in ATTR amyloidosis; however, the findings require confirmation in a randomised controlled trial in patients with ATTR-CM.<sup>40</sup>

The observations above were reinforced by a small, but important study in 16 patients with ATTR-Mixed, treated with patisiran and diflunisal. Treatment was well tolerated and achieved >80% knockdown in serum TTR in 82% of cases which was associated in some patients with a significant reduction in CMR derived ECV and NT-proBNP, and an increase in 6-minute walk (6MWT) test distance following 1 year of treatment. Baseline and follow up bone scintigraphy revealed a reduction in cardiac uptake following patisiran treatment. However, although the reduction in cardiac uptake on bone scintigraphy undeniably demonstrated a favourable biological effect of patisiran, the kinetics and dynamics of radiotracers binding to the myocardium, bones and soft tissues may fluctuate, and therefore



variations in any of these organs will influence both the visual appearance and mathematical calculation of proportionate myocardial radiotracer uptake. Therefore, reduced myocardial uptake on bone scintigraphy should always be supported by improvements in other measures of cardiac function, such as either serum cardiac enzymes or alternative cardiac imaging modalities, before the observed changes are attributed exclusively to a reduction in the burden of cardiac amyloid.<sup>41</sup>

The APOLLO-B trial is a phase III randomised placebo-controlled, double-blind study that enrolled 360 patients, to evaluate the efficacy and safety of patisiran treatment in patients with ATTR-CM. The APOLLO-B trial enrolled patients with both wild-type and variant ATTR-CM who had clinical evidence of heart failure and an elevated NT-proBNP between 300ng/L-8500ng/L. Patients were randomised 1:1 to either patisiran or placebo. Patisiran treatment resulted in sustained reductions in serum TTR, with a mean knockdown of 87% at 12 months. Patisiran demonstrated significant clinical benefit in functional capacity as measured by the change 6-minute walk test (6-MWT) at 12 months compared to placebo, with the decline in 6-MWT in the patisiran group being similar to a typical age-related decline seen in healthy adults. A prespecified sensitivity analysis using a mixed-effects model for repeated measures confirmed the robustness of these findings and demonstrated a significant mean difference in 6-MWT distance at 12 months of 18 meters between the patisiran and placebo groups. Patisiran treatment resulted in a significant and favourable difference in the change from baseline in Kansas City Cardiomyopathy Questionnaire Overall Summary score (KCCQ-OS) at 12 months compared to placebo. However, there was no difference in the time to first event (all-cause hospitalisation, urgent heart failure visits or death). Despite there being no statistically significant differences in mortality between the 2 groups, which may in part reflect the short study duration, the data were numerically in favour of patisiran (4[2%] vs 10[6%]). The efficacy and safety of patisiran treatment in patients with ATTR-CM will continue to be

assessed during the open label extension. It is noteworthy that although the APOLLO-B results have been presented at international conferences, the full manuscript is yet to be published, and therefore more detail is required before formal conclusions can be drawn.<sup>42,43</sup>

### *Safety*

Patisiran is prescribed with premedication consisting of an antihistamine, a corticosteroid, paracetamol and ranitidine to reduce the risk of patients developing infusion related reactions. During the phase 1 trial of patisiran, 21% of participants had mild or moderate infusion related reactions. All mild reactions resolved spontaneously, and moderate reactions resolved after temporarily stopping the infusion and then continuing the infusion at a slower rate.<sup>36</sup> Continued use of patisiran for over 18 months was associated with 62% reduction in serum vitamin A levels, which reflects the physiological function of TTR as a carrier for retinol-binding protein, which in turn, transports vitamin A. Therefore, all patients prescribed patisiran are also routinely prescribed oral vitamin A supplementation (2500 IU per day) as prophylaxis.<sup>31</sup> In the APOLLO phase III trial there were no significant differences in the frequency of adverse events between patients on patisiran treatment and those in the placebo group, and importantly there were no reports of eye complications. Infusion related reactions and peripheral oedema occurred more commonly in the patisiran group, but importantly there was a low incidence of serious adverse events. There were 7 deaths in the patisiran group, none of which were related to treatment, and all were thought to be consistent with the natural progression of ATTRv amyloidosis.<sup>28</sup> In the APOLLO-B trial, patisiran was well tolerated, and the vast majority of adverse events were either mild or moderate in severity. Infusion related reactions, arthralgia and muscle spasms were more common in patients treated with patisiran than in the placebo group. There were 4 deaths in the patisiran group, none of which were deemed to be related to treatment (Table 2).<sup>43</sup>

## **Comparison between patisiran and tafamidis**

Tafamidis remains the only licensed treatment for ATTR-CM, and was approved following the ATTR-ACT trial, which demonstrated a reduction in the composite hierarchical primary endpoint of all-cause mortality and cardiovascular hospitalisations, as well as a lower rate of decline in 6MWT distance and KCCQ-OS score compared with placebo.<sup>25</sup> Despite patisiran treatment resulting in favourable changes across a range of metrics, it has not yet been shown to influence cardiovascular outcomes. Although it is possible that these disparities relate to different drug mechanisms, they are also likely influenced by the differences in study populations and study design. APOLLO-B was a 12 month study and the primary end-point was change in 6MWT, rather than cardiovascular events. ATTR-ACT was a 30 month study and the primary endpoint was a hierarchical composite of all-cause mortality followed by cardiovascular hospitalisations. Baseline patient characteristics indicate more advanced cardiac disease among ATTR-ACT study participants than those in APOLLO-B and consequently the event rate in ATTR-ACT was higher such that a larger population would have been required in APOLLO-B to ensure adequate power to demonstrate a difference in cardiac events. These data also raise questions regarding efficacy of individual treatments at different stages of the disease course, and more data are needed to clarify the relative treatment benefits in different disease stages.<sup>20</sup>

Tafamidis and patisiran differ in their route of administration. Tafamidis is administered orally and is thus convenient; patisiran is an intravenous infusion that requires premedication and co-prescription of prophylactic vitamin A. Both have a good safety profile, although a smaller proportion of patients in the treatment arm of ATTR-ACT reported adverse events than in APOLLO and APOLLO-B, where the majority of events were infusion-related reactions.

However, in the absence of any head-to head trials comparisons between these different treatments are by definition flawed.<sup>25,28,42,43</sup>

Patisiran is able to achieve >80% serum TTR knockdown in the majority of patients. However, this leaves a significant proportion of circulating TTR that could misfold into ATTR fibrils. It is conceivable that simultaneously reducing TTR production and inducing TTR stabilisation could be synergistic and further improve patient outcomes. However, large-scale trials would be needed to assess whether combination therapy has any added benefit.<sup>44</sup>

### **Monitoring the cardiac response to patisiran**

Following promising trial results there has been an increasing interest in use of patisiran to treat ATTR-CM, and at present there is an unmet clinical need to develop standardised measures of the cardiac response to treatment. Advancements in cardiac imaging, combined with increased awareness amongst clinicians has resulted in patients being diagnosed earlier in the disease process; evidenced by a shorter duration of heart failure and neuropathic symptoms prior to diagnosis, and a milder NAC disease stage at the time of diagnosis. This has translated into improved survival and a reduced event rate in the ATTR-CM population, which will invariably have an influence on the design of future clinical trials; with a greater number of patients being recruited and for longer follow-up durations in order to ensure adequate power.<sup>20</sup> Such changes will substantially increase the cost of clinical trials, and the question remains as to whether cardiac biomarkers or imaging measures can be leveraged as surrogates to facilitate a safe but more cost-effective assessment of novel medicines.

NT-proBNP has long been utilised in the heart failure population to assess the response to heart failure medications, and also in cardiac light-chain (AL) amyloidosis to monitor treatment response.<sup>45</sup> However, NT-proBNP fluctuations represent the final common pathway of multiple interlinked mechanisms such as ventricular remodelling, renal dysfunction, fluid

status and neurohormonal activation;<sup>46</sup> hence these changes do not necessarily reflect the change in cardiac amyloid burden.<sup>47</sup> High-sensitivity troponins are widely used in the assessment of multiple acute cardiac conditions,<sup>48</sup> and contribute to risk stratification in cardiac AL amyloidosis.<sup>49</sup> Due to their high-sensitivity, even small fluctuations reflect changes in disease activity.<sup>48</sup> However, their use in monitoring treatment response in ATTR-CM requires further exploration and validation.

The search for a clinical biomarker that can easily measure the cardiac response to patisiran has led to increasing interest in the use of multimodality cardiac imaging. Improvements in various echocardiographic parameters have been reported following treatment with patisiran.<sup>40</sup> However, echocardiographic measures are subject to high levels of intra- and inter-observer variability, which represents a significant limitation when only small changes in echocardiographic parameters are expected.<sup>50</sup> The emergence of data science has the potential to significantly improve precision and overcome these obstacles.<sup>8</sup> Various studies have utilised convolutional neural networks in multiple different modalities of cardiovascular imaging, and to date, all have yielded encouraging results.<sup>51,52</sup> The automation of measurements is associated with a significant improvement in precision, and may facilitate a more accurate assessment of the cardiac response to treatment with patisiran.<sup>51,53</sup>

CMR with multi-parametric mapping has emerging as an essential tool the assessment of ATTR-CM. Amyloidosis is the exemplar interstitial disease, and with amyloid accumulation resulting in expansion of the extracellular matrix. Therefore, myocardial ECV quantification acts as an powerful surrogate measure of the cardiac amyloid burden.<sup>4,54,55</sup> In patients with cardiac AL amyloidosis, changes in ECV following treatment, predate any change in structural or functional parameters, and accurately predict survival even after adjusting for traditional predictors, such as change in NT-proBNP and longitudinal strain.<sup>30</sup> At present, only one small study of patients with ATTR-CM has demonstrated ECV regression following treatment with

patisiran, but if these results were to be reproduced on a larger scale, ECV mapping could become more widely utilised to assess the cardiac response to patisiran (Figure 3).<sup>41,44</sup>

Although reduced cardiac uptake on bone scintigraphy has been reported following treatment with patisiran, changes in myocardial radiotracer uptake are also heavily influenced by changes in the dynamics and kinetics of bone tracers, and therefore bone scintigraphy alone cannot be used as a reliable measure of the cardiac response at present.<sup>41</sup> The use of positron emission tomography (PET) and various novel tracers has shown diagnostic potential in ATTR-CM, but at present there is a lack of robust data supporting their use in tracking treatment response (Table 3).<sup>56-62</sup>

### **Long-term safety of patisiran**

Although short to medium term patisiran-induced TTR knockdown appears safe, studies with prolonged follow-up are required to establish whether there are any long-term side effects. The physiological role of the TTR protein was previously thought to be limited to the transportation of thyroxine and vitamin A. However, various in-vitro and in-vivo studies suggest that TTR may have additional roles within the central nervous system. Animal studies have demonstrated that 5-month-old mice with complete TTR knockdown developed significant memory impairment compared with age matched mice,<sup>63</sup> and TTR deficiency in rats lead to accelerated age-related decline in cognitive performance.<sup>64</sup> These findings have fuelled the hypothesis that the TTR protein may have a role in delaying Alzheimer's related neurodegeneration, a disease process characterised by the accumulation of amyloid- $\beta$  plaques within the brain. In-vitro studies demonstrated that TTR sequesters amyloid- $\beta$  when added to cerebrospinal fluid,<sup>65</sup> and mouse models have demonstrated hippocampal upregulation of TTR in response to the formation of hippocampal amyloid- $\beta$  plaques, suggesting a possible neuroprotective response.<sup>66</sup> Subsequent, anti-TTR antibody infusion led to augmented amyloid- $\beta$  plaque

development, loss of neurones and neuronal apoptosis.<sup>67</sup> In mouse models, surgical occlusion of the middle cerebral artery led to a significantly larger infarct area and greater degree of cerebral oedema in those with complete TTR depletion,<sup>68</sup> while subsequent incubation of the hippocampal neurones with recombinant TTR was associated with improved neurite growth.<sup>69</sup> The possible neuroprotective effect of TTR in cerebrovascular ischaemia has also been observed in humans, with a large observational cohort study of patients with cerebral infarction, demonstrating that serum TTR levels represented an independent predictor of good outcomes.<sup>70</sup> Importantly however, so far there has been no evidence to suggest cognitive decline or abnormality, in patients receiving patisiran treatment continuously for over 5 years.<sup>44</sup> Furthermore, patisiran is unable to cross the blood brain barrier and does not therefore interfere directly with TTR synthesis in the choroid plexus and retinal pigment epithelium, which are responsible for the production of TTR within the cerebrospinal fluid.<sup>71</sup>

### **Future perspectives**

Advances in siRNA conjugate delivery platforms assisted the development of vutrisiran, which is a 3 monthly subcutaneous injection, administered without the need for predication. The HELIOS-A trial randomised ATTRv-PN patients to vutrisiran or patisiran, and compared outcomes to an external placebo group from the APOLLO trial. Vutrisiran treatment achieved similar serum TTR knockdown and was non-inferior to patisiran resulting in improved mNIS+7 and Norfolk QOL-DN scores when compared to placebo. It has since been approved for the treatment of ATTRv-PN.<sup>72,73</sup> The ongoing HELIOS-B trial aims to assess the efficacy of vutrisiran for the treatment of ATTR-CM, and if successful will potentially offer a convenient alternative to patisiran.<sup>74</sup>

### **Conclusion**

ATTR-CM remains a progressive, and ultimately fatal disease. Reduction of the respective circulating APP in all types of amyloidosis in which it is possible has long been known to be associated with improved patient outcomes. Similarly, recent studies have demonstrated that reducing the circulating TTR concentration with patisiran results in improvement in a wide range of cardiac and neurological disease measures including cardiac enzymes, functional capacity, multiple cardiac imaging-based parameters, neurological scores and quality of life. These exciting results will potentially result in expansion of the patisiran license to include ATTR-CM, and increase the armamentarium of disease-modifying treatments available for this aggressive, but treatable cardiomyopathy. Whilst there have been no deleterious clinical consequences in the short to medium term of patisiran-induced TTR depletion, long-term effects remain to be determined.



## References

1. Wechalekar AD, Gillmore JD, Hawkins PN. Systemic amyloidosis. *Lancet* 2016;**387**:2641–2654.
2. Lachmann HJ, Hawkins PN. Systemic amyloidosis. *Curr Opin Pharmacol* 2006;**6**:214–220.
3. Martinez-Naharro A, Hawkins PN, Fontana M. Cardiac amyloidosis. *Clin Med (Lond)* 2018;**18**:s30–s35.
4. Fontana M, Ćorović A, Scully P, Moon JC. Myocardial Amyloidosis: The Exemplar Interstitial Disease. *JACC Cardiovasc Imaging* 2019;**12**:2345–2356.
5. Koike H, Katsuno M. Transthyretin Amyloidosis: Update on the Clinical Spectrum, Pathogenesis, and Disease-Modifying Therapies. *Neurol Ther* 2020;**9**:317.
6. Porcari A, Fontana M, Gillmore JD. Transthyretin cardiac amyloidosis. *Cardiovasc Res* 2022.**118**:3517-3535
7. Patel RK, Ioannou A, Razvi Y, Chacko L, Venneri L, Bandera F, Knight D, Kotecha T, Martinez-Naharro A, Masi A, Porcari A, Brown J, Patel K, Manisty C, Moon J, Rowczenio D, Gilbertson JA, Sinagra G, Lachmann H, Wechalekar A, Petrie A, Whelan C, Hawkins PN, Gillmore JD, Fontana M. Sex differences among patients with transthyretin amyloid cardiomyopathy – from diagnosis to prognosis. *Eur J Heart Fail* 2022.**24**:2355-2363
8. Ioannou A, Patel R, Gillmore JD, Fontana M. Imaging-Guided Treatment for Cardiac Amyloidosis. *Current Cardiology Reports* 2022;**24**:839-850.
9. Ioannou A, Patel RK, Razvi Y, Porcari A, Knight D, Martinez-Naharro A, Kotecha T, Venneri L, Chacko L, Brown J, Manisty C, Moon J, Wisniowski B, Lachmann H, Wechelakar A, Whelan C, Kellman P, Hawkins PN, Gillmore JD, Fontana M. Multi-Imaging Characterization of Cardiac Phenotype in Different Types of Amyloidosis. *JACC Cardiovasc Imaging* 2022.**16**:464-477
10. Martinez-Naharro A, Treibel TA, Abdel-Gadir A, Bulluck H, Zumbo G, Knight DS, Kotecha T, Francis R, Hutt DF, Rezk T, Rosmini S, Quarta CC, Whelan CJ, Kellman P, Gillmore JD, Moon JC, Hawkins PN, Fontana M. Magnetic Resonance in Transthyretin Cardiac Amyloidosis. *J Am Coll Cardiol* 2017;**70**:466–477.
11. Martinez-Naharro A, Gonzalez-Lopez E, Corovic A, Mirelis JG, Baksi AJ, Moon JC, Garcia-Pavia P, Gillmore JD, Hawkins PN, Fontana M. High Prevalence of Intracardiac Thrombi in Cardiac Amyloidosis. *J Am Coll Cardiol* 2019;**73**:1733–1734.
12. Carr AS, Pelayo-Negro AL, Evans MRB, Laurà M, Blake J, Stancanelli C, Iodice V, Wechalekar AD, Whelan CJ, Gillmore JD, Hawkins PN, Reilly MM. A study of the neuropathy associated with transthyretin amyloidosis (ATTR) in the UK. *J Neurol Neurosurg Psychiatry* 2016;**87**:620–627.
13. Yanagisawa A, Ueda M, Sueyoshi T, Okada T, Fujimoto T, Ogi Y, Kitagawa K, Tasaki M, Misumi Y, Oshima T, Jono H, Obayashi K, Hirakawa K, Uchida H, Westermarck P, Ando Y, Mizuta H. Amyloid deposits derived from transthyretin in the ligamentum flavum as related to lumbar spinal canal stenosis. *Mod Pathol* 2015;**28**:201–207.
14. Geller HI, Singh A, Alexander KM, Mirto TM, Falk RH. Association Between Ruptured Distal Biceps Tendon and Wild-Type Transthyretin Cardiac Amyloidosis. *JAMA* 2017;**318**:962–963.
15. Koike H, Okumura T, Murohara T, Katsuno M. Multidisciplinary Approaches for Transthyretin Amyloidosis. *Cardiol Ther* 2021;**10**:289–311.

16. Arruda-Olson AM, Zeldenrust SR, Dispenzieri A, Gertz MA, Miller FA, Bielinski SJ, Klarich KW, Scott CG, Grogan M. Genotype, echocardiography, and survival in familial transthyretin amyloidosis. *Amyloid* 2013;**20**:263–268.
17. Porcari A, Razvi Y, Masi A, Patel R, Ioannou A, Rauf MU, Hutt DF, Rowczenio D, Gilbertson J, Martinez-Naharro A, Venneri L, Whelan C, Lachmann H, Wechalekar A, Quarta CC, Merlo M, Sinagra G, Hawkins PN, Fontana M, Gillmore JD. Prevalence, Characteristics and Outcomes of Older Patients with Hereditary versus Wild-Type Transthyretin Amyloid Cardiomyopathy. *Eur J Heart Fail* 2023.
18. Gillmore JD, Maurer MS, Falk RH, Merlini G, Damy T, Dispenzieri A, Wechalekar AD, Berk JL, Quarta CC, Grogan M, Lachmann HJ, Bokhari S, Castano A, Dorbala S, Johnson GB, Glaudemans AWJM, Rezk T, Fontana M, Palladini G, Milani P, Guidalotti PL, Flatman K, Lane T, Vonberg FW, Whelan CJ, Moon JC, Ruberg FL, Miller EJ, Hutt DF, Hazenberg BP, Rapezzi C, Hawkins PN. Nonbiopsy Diagnosis of Cardiac Transthyretin Amyloidosis. *Circulation* 2016;**133**:2404–2412.
19. Rauf MU, Hawkins PN, Cappelli F, Perfetto F, Zampieri M, Argiro A, Petrie A, Law S, Porcari A, Razvi Y, Bomszyk J, Ravichandran S, Ioannou A, Patel R, Starr N, Hutt DF, Mahmood S, Wisniowski B, Martinez-Naharro A, Venneri L, Whelan C, Rowczenio D, Gilbertson J, Lachmann HJ, Wechalekar AD, Rapezzi C, Serenelli M, Massa P, Caponetti AG, Ponziani A, Accietto A, Giovannetti A, Sauri G, Sguazzotti M, Gagliardi C, Biagini E, Longhi S, Fontana M, Gillmore JD. Tc-99m labelled bone scintigraphy in suspected cardiac amyloidosis. *Eur Heart J* 2023;**00**:1–12.
20. Ioannou A, Patel RK, Razvi Y, Porcari A, Sinagra G, Venneri L, Bandera F, Masi A, Williams GE, O’Beara S, Ganesanathan S, Massa P, Knight D, Martinez-Naharro A, Kotecha T, Chacko L, Brown J, Rauf MU, Manisty C, Moon J, Lachmann H, Wechalekar A, Petrie A, Whelan C, Hawkins PN, Gillmore JD, Fontana M. Impact of Earlier Diagnosis in Cardiac ATTR Amyloidosis Over the Course of 20 Years. *Circulation* 2022;**29**:1657-1670
21. Lauppe RE, Liseth Hansen J, Gerdesköld C, Rozenbaum MH, Strand AM, Vakevainen M, Kuusisto J, Gude E, Gustafsson F, Gustav Smith J. Original research: Nationwide prevalence and characteristics of transthyretin amyloid cardiomyopathy in Sweden. *Open Heart* 2021;**8**:1755.
22. Tanskanen M, Peuralinna T, Polvikoski T, Notkola IL, Sulkava R, Hardy J, Singleton A, Kiuru-Enari S, Paetau A, Tienari PJ, Myllykangas L. Senile systemic amyloidosis affects 25% of the very aged and associates with genetic variation in alpha2-macroglobulin and tau: a population-based autopsy study. *Ann Med* 2008;**40**:232–239.
23. Adams D, Algalarrondo V, Polydefkis M, Sarswat N, Slama MS, Nativi-Nicolau J. Expert opinion on monitoring symptomatic hereditary transthyretin-mediated amyloidosis and assessment of disease progression. *Orphanet J Rare Dis* 2021;**16**:411.
24. Carvalho A, Rocha A, Lobato L. Liver transplantation in transthyretin amyloidosis: issues and challenges. *Liver Transpl* 2015;**21**:282–292.
25. Maurer MS, Schwartz JH, Gundapaneni B, Elliott PM, Merlini G, Waddington-Cruz M, Kristen A V., Grogan M, Witteles R, Damy T, Drachman BM, Shah SJ, Hanna M, Judge DP, Barsdorf AI, Huber P, Patterson TA, Riley S, Schumacher J, Stewart M, Sultan MB, Rapezzi C. Tafamidis Treatment for Patients with Transthyretin Amyloid Cardiomyopathy. *N Engl J Med* 2018;**379**:1007–1016.
26. Lachmann HJ, Goodman HJB, Gilbertson JA, Gallimore JR, Sabin CA, Gillmore JD, Hawkins PN. Natural history and outcome in systemic AA amyloidosis. *N Engl J Med* 2007;**356**:2361–2371.
27. Palladini G, Dispenzieri A, Gertz MA, Kumar S, Wechalekar A, Hawkins PN, Schönland S, Heggenbart U, Comenzo R, Kastiris E, Dimopoulos MA, Jaccard A,

- Klersy C, Merlini G. New criteria for response to treatment in immunoglobulin light chain amyloidosis based on free light chain measurement and cardiac biomarkers: Impact on survival outcomes. *Journal of Clinical Oncology* 2012;**30**:4541–4549.
28. Adams D, Gonzalez-Duarte A, O’Riordan WD, Yang C-C, Ueda M, Kristen A V., Tournev I, Schmidt HH, Coelho T, Berk JL, Lin K-P, Vita G, Attarian S, Planté-Bordeneuve V, Mezei MM, Campistol JM, Buades J, Brannagan TH, Kim BJ, Oh J, Parman Y, Sekijima Y, Hawkins PN, Solomon SD, Polydefkis M, Dyck PJ, Gandhi PJ, Goyal S, Chen J, Strahs AL, Nochur S V., Sweetser MT, Garg PP, Vaishnav AK, Gollob JA, Suhr OB. Patisiran, an RNAi Therapeutic, for Hereditary Transthyretin Amyloidosis. *N Engl J Med* 2018;**379**:11–21.
  29. Hawkins PN, Lavender JP, Pepys MB. Evaluation of systemic amyloidosis by scintigraphy with <sup>123</sup>I-labeled serum amyloid P component. *N Engl J Med* 1990;**323**:508–513.
  30. Martinez-Naharro A, Patel R, Kotecha T, Karia N, Ioannou A, Petrie A, Chacko LA, Razvi Y, Ravichandran S, Brown J, Law S, Quarta C, Mahmood S, Wisniowski B, Pica S, Sachchithanatham S, Lachmann HJ, Moon JC, Knight DS, Whelan C, Venneri L, Xue H, Kellman P, Gillmore JD, Hawkins PN, Wechalekar AD, Fontana M. Cardiovascular magnetic resonance in light-chain amyloidosis to guide treatment. *Eur Heart J* 2022;**1**:4722-4735.
  31. Urits I, Swanson D, Swett MC, Patel A, Berardino K, Amgalan A, Berger AA, Kassem H, Kaye A, Viswanath O. A Review of Patisiran (ONPATRO®) for the Treatment of Polyneuropathy in People with Hereditary Transthyretin Amyloidosis. *Neurol Ther* 2020;**9**:301–315.
  32. Setten RL, Rossi JJ, Han S ping. The current state and future directions of RNAi-based therapeutics. *Nat Rev Drug Discov* 2019;**18**:421–446.
  33. Hu B, Zhong L, Weng Y, Peng L, Huang Y, Zhao Y, Liang XJ. Therapeutic siRNA: state of the art. *Signal Transduct Target Ther* 2020;**5**.
  34. Caplen NJ, Mousset S. Short interfering RNA (siRNA)-mediated RNA interference (RNAi) in human cells. *Ann N Y Acad Sci* 2003;**1002**:56–62.
  35. Friedrich M, Aigner A. Therapeutic siRNA: State-of-the-Art and Future Perspectives. 123AD.
  36. Coelho T, Adams D, Silva A, Lozeron P, Hawkins PN, Mant T, Perez J, Chiesa J, Warrington S, Tranter E, Munisamy M, Falzone R, Harrop J, Cehelsky J, Bettencourt BR, Geissler M, Butler JS, Sehgal A, Meyers RE, Chen Q, Borland T, Hutabarat RM, Clausen VA, Alvarez R, Fitzgerald K, Gamba-Vitalo C, Nochur S v., Vaishnav AK, Sah DWY, Gollob JA, Suhr OB. Safety and Efficacy of RNAi Therapy for Transthyretin Amyloidosis. *New England Journal of Medicine* 2013;**369**:819–829.
  37. Suhr OB, Coelho T, Buades J, Pouget J, Conceicao I, Berk J, Schmidt H, Waddington-Cruz M, Campistol JM, Bettencourt BR, Vaishnav A, Gollob J, Adams D. Efficacy and safety of patisiran for familial amyloidotic polyneuropathy: a phase II multi-dose study. *Orphanet J Rare Dis* 2015;**10**.
  38. Coelho T, Adams D, Conceição I, Waddington-Cruz M, Schmidt HH, Buades J, Campistol J, Berk JL, Polydefkis M, Wang JJ, Chen J, Sweetser MT, Gollob J, Suhr OB. A phase II, open-label, extension study of long-term patisiran treatment in patients with hereditary transthyretin-mediated (hATTR) amyloidosis. *Orphanet J Rare Dis* 2020;**15**.
  39. González-Duarte A, Berk JL, Quan D, Mauermann ML, Schmidt HH, Polydefkis M, Waddington-Cruz M, Ueda M, Conceição IM, Kristen A v., Coelho T, Cauquil CA, Tard C, Merkel M, Aldinc E, Chen J, Sweetser MT, Wang JJ, Adams D. Analysis of autonomic outcomes in APOLLO, a phase III trial of the RNAi therapeutic patisiran in

- patients with hereditary transthyretin-mediated amyloidosis. *J Neurol* 2020;**267**:703–712.
40. Solomon SD, Adams D, Kristen A, Grogan M, González-Duarte A, Maurer MS, Merlini G, Damy T, Slama MS, Brannagan TH, Dispenzieri A, Berk JL, Shah AM, Garg P, Vaishnav A, Karsten V, Chen J, Gollob J, Vest J, Suhr O. Effects of Patisiran, an RNA Interference Therapeutic, on Cardiac Parameters in Patients With Hereditary Transthyretin-Mediated Amyloidosis. *Circulation* 2019;**139**:431–443.
  41. Fontana M, Martinez-Naharro A, Chacko L, Rowczenio D, Gilbertson JA, Whelan CJ, Strehina S, Lane T, Moon J, Hutt DF, Kellman P, Petrie A, Hawkins PN, Gillmore JD. Reduction in CMR Derived Extracellular Volume With Patisiran Indicates Cardiac Amyloid Regression. *JACC Cardiovasc Imaging* 2021;**14**:189–199.
  42. Topline Results from APOLLO-B Phase 3 Study of Patisiran 2 Agenda Welcome. 2022.
  43. Maurer MS, Fontana M, Berk JL, Gustafsson F, Simões M, Grogan M, Fernandes F, Gottlieb RL, Kubanek M, Poulsen S. Primary Results from APOLLO-B, A Phase 3 Study of Patisiran in Patients with Transthyretin-Mediated Amyloidosis with Cardiomyopathy
  44. Ioannou A, Fontana M, Gillmore JD. RNA Targeting and Gene Editing Strategies for Transthyretin Amyloidosis. *BioDrugs* 2023;**37**:127-142
  45. Merlini G, Lousada I, Ando Y, Dispenzieri A, Gertz MA, Grogan M, Maurer MS, Sanchorawala V, Wechalekar A, Palladini G, Comenzo RL. Rationale, application and clinical qualification for NT-proBNP as a surrogate end point in pivotal clinical trials in patients with AL amyloidosis. *Leukemia* 2016;**30**:1979–1986.
  46. Emdin M, Passino C, Prontera C, Iervasi A, Ripoli A, Masini S, Zucchelli GC, Clerico A. Cardiac natriuretic hormones, neuro-hormones, thyroid hormones and cytokines in normal subjects and patients with heart failure. *Clin Chem Lab Med* 2004;**42**:627–636.
  47. Banypersad SM, Sado DM, Flett AS, Gibbs SDJ, Pinney JH, Maestrini V, Cox AT, Fontana M, Whelan CJ, Wechalekar AD, Hawkins PN, Moon JC. Quantification of myocardial extracellular volume fraction in systemic AL amyloidosis: an equilibrium contrast cardiovascular magnetic resonance study. *Circ Cardiovasc Imaging* 2013;**6**:34–39.
  48. Passino C, Aimò A, Masotti S, Musetti V, Prontera C, Emdin M, Clerico A. Cardiac troponins as biomarkers for cardiac disease. *Biomark Med* 2019;**13**:325–330.
  49. Dispenzieri A, Gertz MA, Kumar SK, Lacy MQ, Kyle RA, Saenger AK, Grogan M, Zeldenrust SR, Hayman SR, Buadi F, Greipp PR, Leu GN, Russell SR, Dingli D, Lust JA, Rajkumar S v., Jaffe AS. High sensitivity cardiac troponin T in patients with immunoglobulin light chain amyloidosis. *Heart* 2014;**100**:383–388.
  50. Chacko L, Karia N, Venneri L, Bandera F, Passo BD, Buonamici L, Lazari J, Ioannou A, Porcari A, Patel R, Razvi Y, Brown J, Knight D, Martinez-Naharro A, Whelan C, Quarta CC, Manisty C, Moon J, Rowczenio D, Gilbertson JA, Lachmann H, Wechelakar A, Petrie A, Moody WE, Steeds RP, Potena L, Riefolo M, Leone O, Rapezzi C, Hawkins PN, Gillmore JD, Fontana M. Progression of echocardiographic parameters and prognosis in transthyretin cardiac amyloidosis. *Eur J Heart Fail* 2022;**24**:1700-1712.
  51. Dey D, Slomka PJ, Leeson P, Comaniciu D, Shrestha S, Sengupta PP, Marwick TH. Artificial Intelligence in Cardiovascular Imaging: JACC State-of-the-Art Review. *J Am Coll Cardiol* 2019;**73**:1317–1335.
  52. Tromp J, Seekings PJ, Hung CL, Iversen MB, Frost MJ, Ouwerkerk W, Jiang Z, Eisenhaber F, Goh RSM, Zhao H, Huang W, Ling LH, Sim D, Cozzone P, Richards AM, Lee HK, Solomon SD, Lam CSP, Ezekowitz JA. Automated interpretation of

- systolic and diastolic function on the echocardiogram: a multicohort study. *Lancet Digit Health* 2022;**4**:e46–e54.
53. Howard JP, Francis DP. Machine learning with convolutional neural networks for clinical cardiologists. *Heart* 2021;**108**:973–981.
  54. Fontana M, Banypersad SM, Treibel TA, Maestrini V, Sado DM, White SK, Pica S, Castelletti S, Piechnik SK, Robson MD, Gilbertson JA, Rowczenio D, Hutt DF, Lachmann HJ, Wechalekar AD, Whelan CJ, Gillmore JD, Hawkins PN, Moon JC. Native T1 mapping in transthyretin amyloidosis. *JACC Cardiovasc Imaging* 2014;**7**:157–165.
  55. Karamitsos TD, Piechnik SK, Banypersad SM, Fontana M, Ntusi NB, Ferreira VM, Whelan CJ, Myerson SG, Robson MD, Hawkins PN, Neubauer S, Moon JC. Noncontrast T1 mapping for the diagnosis of cardiac amyloidosis. *JACC Cardiovasc Imaging* 2013;**6**:488–497.
  56. Dorbala S, Vangala D, Semer J, Strader C, Bruyere JR, Carli MF di, Moore SC, Falk RH. Imaging cardiac amyloidosis: a pilot study using <sup>18</sup>F-florbetapir positron emission tomography. *Eur J Nucl Med Mol Imaging* 2014;**41**:1652–1662.
  57. Dietemann S, Nkoulou R. Amyloid PET imaging in cardiac amyloidosis: a pilot study using 18 F-flutemetamol positron emission tomography. *Ann Nucl Med* 2019;**33**:624–628.
  58. Antoni G, Lubberink M, Estrada S, Axelsson J, Carlson K, Lindsjö L, Kero T, Långström B, Granstam SO, Rosengren S, Vedin O, Wassberg C, Wikström G, Westermark P, Sörensen J. In vivo visualization of amyloid deposits in the heart with 11C-PIB and PET. *J Nucl Med* 2013;**54**:213–220.
  59. Genovesi D, Vergaro G, Giorgetti A, Marzullo P, Scipioni M, Santarelli MF, Pucci A, Buda G, Volpi E, Emdin M. [18F]-Florbetaben PET/CT for Differential Diagnosis Among Cardiac Immunoglobulin Light Chain, Transthyretin Amyloidosis, and Mimicking Conditions. *JACC Cardiovasc Imaging* 2021;**14**:246–255.
  60. Kircher M, Ihne S, Brumberg J, Morbach C, Knop S, Kortüm KM, Störk S, Buck AK, Reiter T, Bauer WR, Lapa C. Detection of cardiac amyloidosis with 18 F-Florbetaben-PET/CT in comparison to echocardiography, cardiac MRI and DPD-scintigraphy. *Eur J Nucl Med Mol Imaging* 2019;**46**:1407–1416.
  61. Lee SP, Suh HY, Park S, Oh S, Kwak SG, Kim HM, Koh Y, Park JB, Kim HK, Cho HJ, Kim YJ, Kim I, Yoon SS, Seo JW, Paeng JC, Sohn DW. Pittsburgh B Compound Positron Emission Tomography in Patients With AL Cardiac Amyloidosis. *J Am Coll Cardiol* 2020;**75**:380–390.
  62. Kim YJ, Ha S, Kim Y il. Cardiac amyloidosis imaging with amyloid positron emission tomography: A systematic review and meta-analysis. *J Nucl Cardiol* 2020;**27**:123–132.
  63. Sousa JC, Marques F, Dias-Ferreira E, Cerqueira JJ, Sousa N, Palha JA. Transthyretin influences spatial reference memory. *Neurobiol Learn Mem* 2007;**88**:381–385.
  64. Brouillette J, Quirion R. Transthyretin: a key gene involved in the maintenance of memory capacities during aging. *Neurobiol Aging* 2008;**29**:1721–1732.
  65. Schwarzman AL, Gregori L, Vitek MP, Lyubski S, Strittmatter WJ, Enghilde JJ, Bhasin R, Silverman J, Weisgraber KH, Coyle PK, Zagorski MG, Talafous J, Eisenberg M, Saunders AM, Roses AD, Goldgaber D. Transthyretin sequesters amyloid beta protein and prevents amyloid formation. *Proc Natl Acad Sci U S A* 1994;**91**:8368.
  66. Stein TD, Johnson JA. Lack of neurodegeneration in transgenic mice overexpressing mutant amyloid precursor protein is associated with increased levels of transthyretin and the activation of cell survival pathways. *J Neurosci* 2002;**22**:7380–7388.

67. Stein TD, Anders NJ, DeCarli C, Chan SL, Mattson MP, Johnson JA. Neutralization of transthyretin reverses the neuroprotective effects of secreted amyloid precursor protein (APP) in APPSW mice resulting in tau phosphorylation and loss of hippocampal neurons: support for the amyloid hypothesis. *J Neurosci* 2004;**24**:7707–7717.
68. Santos SD, Lambertsen KL, Clausen BH, Akinc A, Alvarez R, Finsen B, Saraiva MJ. CSF transthyretin neuroprotection in a mouse model of brain ischemia. *J Neurochem* 2010;**115**:1434–1444.
69. Gomes JR, Nogueira RS, Vieira M, Santos SD, Ferraz-Nogueira JP, Relvas JB, Saraiva MJ. Transthyretin provides trophic support via megalin by promoting neurite outgrowth and neuroprotection in cerebral ischemia. *Cell Death Differ* 2016;**23**:1749.
70. Gao C, Zhang B, Zhang W, Pu S, Yin J, Gao Q. Serum prealbumin (transthyretin) predict good outcome in young patients with cerebral infarction. *Clin Exp Med* 2011;**11**:49–54.
71. Luigetti M, Servidei S. Patisiran in hereditary transthyretin-mediated amyloidosis. *Lancet Neurol* 2021;**20**:21–23.
72. Adams D, Tournev IL, Taylor MS, Coelho T, Planté-Bordeneuve V, Berk JL, González-Duarte A, Gillmore JD, Low SC, Sekijima Y, Obici L, Chen C, Badri P, Arum SM, Vest J, Polydefkis M. Efficacy and safety of vutrisiran for patients with hereditary transthyretin-mediated amyloidosis with polyneuropathy: a randomized clinical trial. *Amyloid* 2022;**30**:1-9.
73. Keam SJ. Vutrisiran: First Approval. *Drugs* 2022 2022:1–7.
74. HELIOS-B: A Study to Evaluate Vutrisiran in Patients With Transthyretin Amyloidosis With Cardiomyopathy - Full Text View - ClinicalTrials.gov <https://www.clinicaltrials.gov/ct2/show/NCT04153149> (12 September 2022)

## Figure legends

Figure 1. Characteristic echocardiographic features of ATTR cardiomyopathy. A = Parasternal long axis view demonstrating increased interventricular and inferolateral wall thickness. B = Parasternal short axis view demonstrating increased wall thickness. C = 4-chamber view demonstrating biventricular increased wall thickness. D = Pulsed wave doppler of the mitral valve demonstrating an E:A > 2 and grade 3 diastolic dysfunction. E = Longitudinal strain (LS) measured in the 4-chamber view. F = Increased septal apical to basal strain ratio. G-H = Longitudinal strain map demonstrating relative apical sparing and a 'bull's eye' strain pattern.

Figure 2. Characteristic cardiac magnetic resonance imaging features of ATTR cardiomyopathy. Cine images demonstrate asymmetrical septal increased wall thickness and an increased left ventricular mass. Tissue characterisation demonstrates a global elevation in myocardial native T1, biventricular circumferential late gadolinium enhancement and a global elevation in myocardial extracellular volume (ECV).

Figure 3. Cardiac magnetic resonance images demonstrating a reduction in the amount of late gadolinium enhancement and a reduction in the extracellular volume following 3 years of treatment with patisiran.