Diagnosis of ATTR

MED-US-DZSTATE-2400018

ATTR Disease State Slide Deck

- This resource provides information about ATTR.
- This resource is intended to be viewed in its entirety to support scientific exchange and is not intended as recommendations for clinical practice.
- This resource may contain hyperlinks that are not functional in this format.
- For further information, please see RNAiScience.com to connect with a Medical Science Liaison, submit a medical information request, or access other Alnylam medical education resources.

Alnylam®

Diagnosis

Alnylam®

Red-flag Symptoms of ATTR-PN

Progressive symmetric sensorimotor neuropathy

+

≥1 of

Family history
Early autonomic dysfunction (eg, erectile dysfunction or postural hypotension)
GI complaints (eg, chronic diarrhea or constipation)
Unexplained weight loss
Cardiac hypertrophy, arrhythmias, ventricular blocks, or cardiomyopathy
Bilateral CTS (especially if also present in family members)
Renal abnormalities (eg, albuminuria or mild azotemia)
Vitreous opacities

ATTR-PN, ATTR with polyneuropathy; CTS, carpal tunnel syndrome; GI, gastrointestinal.

1. Conceição et al. *J Peripher Nerv Syst.* 2016;21:5–9.

Small and Large Fiber–Associated Sensory, Autonomic, and Motor Dysfunction Throughout the Progression of ATTR-PN[1]

Small fiber[1-3]

Associated with presentation of sensory and autonomic dysfunction: paresthesia, allodynia in the feet, sensory loss of pain and temperature sensation, GI disturbances, and sexual dysfunction

Diagnostic tests[1,4,5]

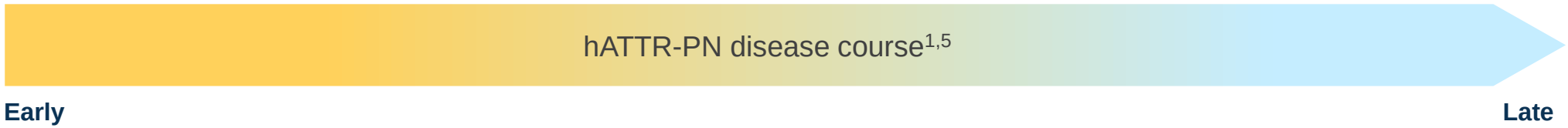
- Skin biopsy
- QSART
- QST

Large fiber[1,2]

Associated with impaired vibration sensation and proprioception, and weakness that starts in the feet and hands but progresses proximally

Diagnostic tests[1]

- NCS
- Needle electromyogram
- Nerve ultrasound



ATTR-PN, ATTR with polyneuropathy; hATTR, hereditary ATTR with polyneuropathy; wtATTR, wild-type ATTR; GI, gastrointestinal; NCS, nerve conduction studies; QSART, quantitative sudomotor axon reflex test; QST, quantitative sensory testing.

1. Namiranian and Geisler. *Am J Med.* 2022;135 Suppl 1:S13–19. 2. Kaku and Berk. *Semin Neurol.* 2019;39:578–88; 3. Papagianni et al. *Amyloid.* 2022;29(1):14–22; 4. Shy et al. *Neurology.* 2003;60:898–904; 5. Shin et al. *Mt Sinai J Med.* 2012;79(6):733–48.

Small and Large Fiber Diagnostic Findings

Neuropathy	Diagnostic test	Findings
Small fiber	Skin biopsy[1]	• Reduced intraepidermal nerve fibers • Variable rates of amyloid detection which correlate with neuropathy severity in ATTR
	QSART[2]	• Reduced or absent sweat response
	QST[3]	• Abnormal thresholds to cold and vibration
Large fiber	Nerve conduction study[1]	Predominant axonal pathology: • Decreased/absent sensory nerve action potential amplitudes • Decreased compound muscle action potential amplitudes • Normal/slight reduction in conduction velocity[a]
	Needle electromyogram[1]	• Fibrillations and positive sharp waves • Neurogenic motor unit potentials (MUPs): broad duration, large amplitude potentials with reduced recruitment
	Nerve ultrasound[1]	• Increased cross-sectional areas in common entrapment sites and in proximal nerve segments

[a]Several reports of amyloid neuropathies with conduction velocities in the demyelinating range have been published and are a major source of misdiagnosis as acquired demyelinating neuropathies such as CIDP.
ATTR, transthyretin-mediated; hATTR, hereditary transthyretin-mediated; wtATTR, wild-type transthyretin-mediated; MUP, motor unit potential; QSART, quantitative sudomotor axon reflex test; QST, quantitative sensory testing.
1. Namiranian and Geisler. *Am J Med.* 2022;135 Suppl 1:S13–19. 2. Shin et al. *Mt Sinai J Med.* 2012;79(6):733–48; 3. Shy et al. *Neurology.* 2003;60:898–904.

Red-flag Symptoms of ATTR-CM

Reduction in longitudinal strain with apical sparing

Discrepancy between LV thickness and QRS voltage (with a lack of LV hypertrophy on ECG)

Atrioventricular block, in the presence of increased LV wall thickness

Echocardiographic hypertrophic phenotype with associated infiltrative features, including increased thickness of the atrioventricular valves, interatrial septum, and RV free wall

Marked extracellular volume expansion, abnormal nulling time for the myocardium, or diffuse late gadolinium enhancement on CMR

Symptoms of polyneuropathy and/or dysautonomia

History of bilateral CTS

Mild increase in troponin levels on repeated occasions

ATTR-CM, ATTR with cardiomyopathy; CMR, cardiac magnetic resonance; CTS, carpal tunnel syndrome; ECG, electrocardiogram; LV, left ventricle; RV, right ventricle.

1. Witteles et al. *J Am Coll Cardiol HF.* 2019;7:709–16.

Alnylam®

Diagnostic Algorithm for Suspected hATTR-PN

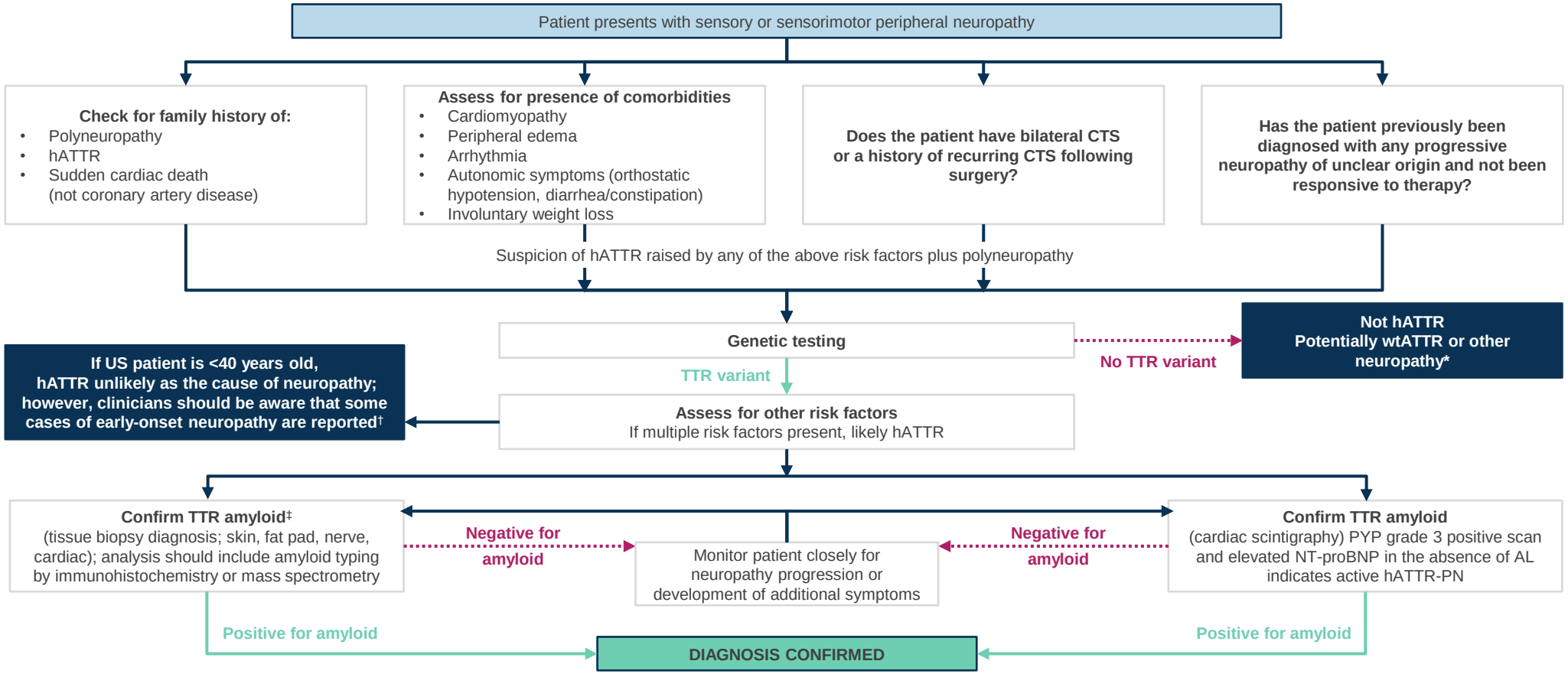


Figure adapted from Karam et al. 2024[1]

*Patients may be assessed for genetic conditions including Charcot–Marie–Tooth disease and hereditary neuropathy with liability to pressure palsies, or screened for vitamin B12 deficiency, diabetes (hemoglobin A1C assessment), thyroid dysfunction, monoclonal gammopathy (immunofixation electrophoresis), or AL amyloidosis (immunoglobulin free light chain assessment).
†Early onset of polyneuropathy has been reported in hATTR.
‡Importance of tissue diagnosis is greater when concurrent possible causes of peripheral neuropathy (ie, B12 deficiency, diabetes mellitus, paraproteinemia, etc) are present. In certain cases where there is no alternative cause for a progressive neuropathy, especially when multisystem features are present, a biopsy may not be necessary. A negative tissue biopsy in a patient with a high suspicion of hATTR does not exclude a diagnosis, and further investigation (i.e., scintigraphy) or close follow-up is warranted.
AL, amyloid light chain; CTS, carpal tunnel syndrome; hATTR, hereditary ATTR with polyneuropathy; NT-proBNP, N-terminal pro-brain natriuretic peptide; PYP, pyrophosphate; TTR, transthyretin; US, United States; wtATTR, wild-type ATTR.
1. Karam et al. *Muscle Nerve.* 2024; S9(3) 273-297.

ACC Diagnostic Algorithm for Suspected Cardiac Amyloidosis

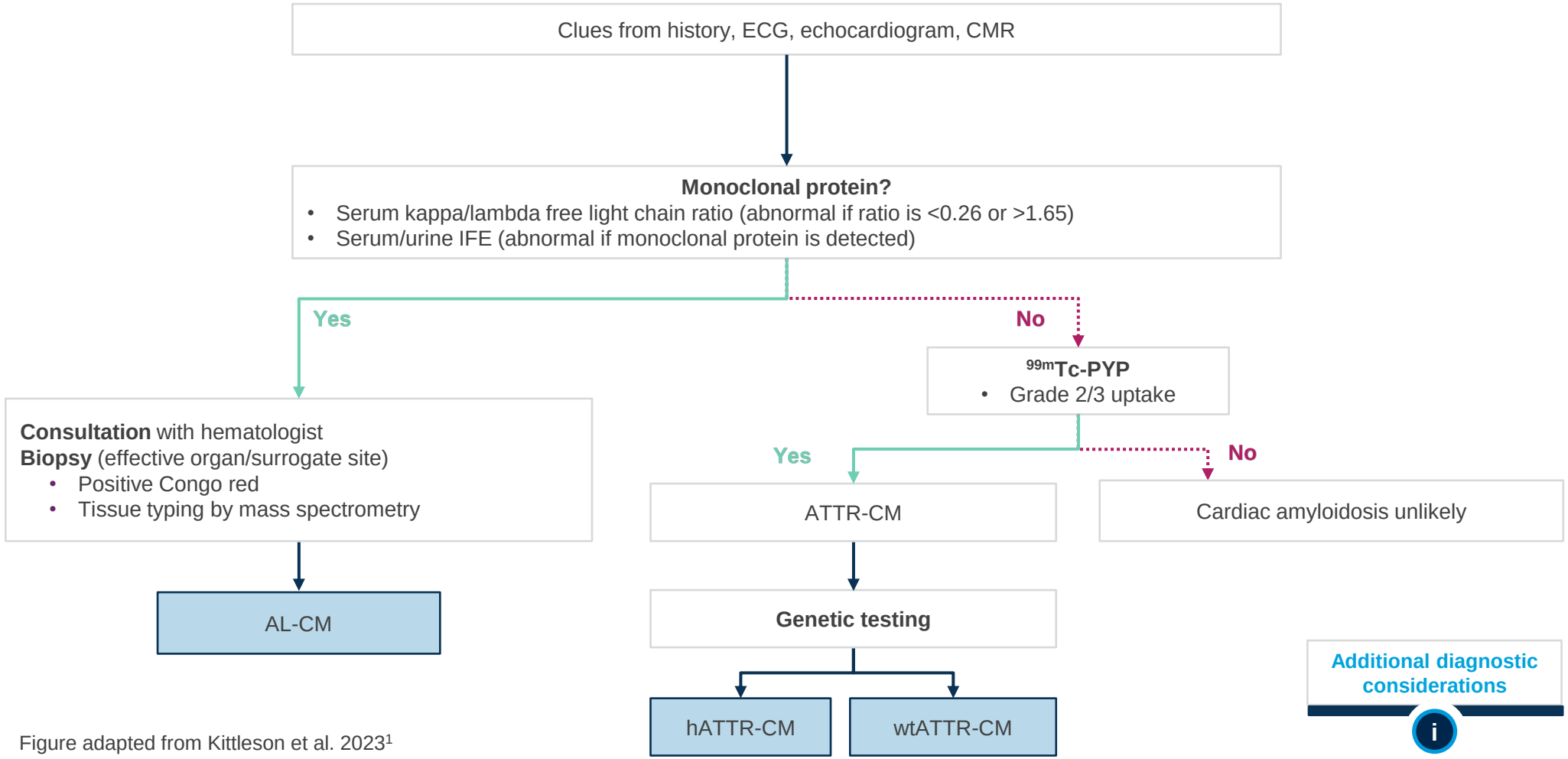


Figure adapted from Kittleson et al. 2023[1]

ACC, American College of Cardiology; AL-CM, primary/amyloid light chain cardiac amyloidosis; ATTR-CM, ATTR with cardiomyopathy; CMR, cardiac magnetic resonance; ECG, electrocardiogram; hATTR, hereditary ATTR; hATTR-CM, hereditary ATTR with cardiomyopathy; ^{99m}Tc-PYP, ^{99m} technetium pyrophosphate; SPECT, single-photon emission computed tomography; wtATTR, wild-type ATTR; wtATTR-CM, wild-type ATTR with cardiomyopathy.

1. Kittleson et al. *JACC.* 2023; 81(11):1076–176.

Considerations During Various Stages of Cardiac Amyloidosis Diagnosis

Monoclonal protein analysis

- SPEP/UPEP not as sensitive as IFE
- Normal K/L ratio in severe kidney disease: 0.54-3.30

Biopsy test

- Surrogate site (fat pad) lacks sensitivity

^{99m}Tc-PYP scintigraphy

- Cardiac scintigraphy could be ordered simultaneously for efficiency but must be interpreted in the context of the negative monoclonal protein screen
- Avoid false positives: use of SPECT imaging to exclude blood pool uptake
- Avoid false negatives: consider biopsy if scintigraphy scan is negative/equivocal but clinical suspicion is high

IFE, immunofixation electrophoresis; K/L, kappa/lambda, ^{99m}Tc-PYP, ^{99m} technetium pyrophosphate; SPECT, single-photon emission computed tomography; SPEP/UPEP, serum/urine protein electrophoresis.

1. Kittleson et al. *JACC.* 2023; 81(11):1076–176.

Non-confirmatory Diagnostic Tools for ATTR

Neuropathy assessments[1-9]

- EMG/Nerve conduction study
- QST
- Sudoscan and QSART
- Autonomic testing

Cardiomyopathy assessments[10-14]

- ECG
- Echocardiogram
- Cardiac MRI

i = Additional info

ATTR, transthyretin amyloidosis; ECG, electrocardiogram; EMG, electromyography; MRI, magnetic resonance imaging; QSART, quantitative sudomotor axon reflex test; QST, quantitative sensory testing.

1. Shy et al. *Neurology.* 2003;60:898–904; 2. Suanprasert et al. *J Neurol Sci.* 2014;344:121–8; 3. Shin & Robinson-Papp. *Mt Sinai J Med.* 2012;79:733–48; 4. Adams et al. *Rev Neurol. (Paris)* 2016;172:645–52; 5. Vaxman et al. *Acta Haematol. 2020;143(4):304-311*; 6. Namiranian et al. *Am J Med.* 2022;135 Suppl 1:S13–19; 7. Illigens & Gibbons. *Clin Auton Res.* 2009;19:79–87; 8. Shields. *Cleve Clin J Med* 2009;76(Suppl 2): S37–40; 9. Teodorovich and Swissa. *World J Cardiol.* 2016;8(3):277–82; 10. Ruberg & Berk. *Circulation.* 2012;126:1286–300; 11. Dharmarajan et al. *J Am Geriatr Soc.* 2012;60:765–74; 12. Gertz et al. *BMC Fam Pract.* 2020;21:198; 13. Castano et al. *Curr Cardiovasc Risk Rep* 2017;11:17; 14. Phelan et al. *Heart.* 2012;98:1442–8.

Alnylam®

Electromyography (EMG)/Nerve Conduction Study (NCS)[1-4]

Neuropathy assessments

- EMG measures **electrical activity in muscles**, while NCS measures **how quickly and effectively nerves send electrical signals**
  - Can be used to assess the degree of neurologic damage and **monitor progression** after diagnosis
- EMG may not detect neuropathy when only small fibers are involved, commonly during early disease
- EMG and NCS are useful in demonstrating large fiber neuropathy in sufficiently advanced cases

EMG, electromyography; NCS, nerve conduction study.

1. Shin & Robinson-Papp. *Mt Sinai J Med.* 2012;79:733–48; 2. Adams et al. *Rev Neurol. (Paris)* 2016;172:645–52; 3. Vaxman et al. *Acta Haematol. 2020;143(4):304-311*; 4. Namiranian et al. *Am J Med.* 2022;135 Suppl 1:S13–19.

Quantitative Sensory Testing (QST)[1,2]

Neuropathy assessment

- **Detect damage to nerve endings** that are used to detect temperature and vibration
- Used to establish the involvement in small sensory nerve fibers in a neuropathy

QST, quantitative sensory testing.
1. Shy et al. *Neurology.* 2003;60:898–904; 2. Suanprasert et al. *J Neurol Sci.* 2014;344:121–8.

Sudoscan/Quantitative Sudomotor Axon Reflex Test (QSART)[1,2]

Neuropathy assessments

- Measure sweat gland function (sudomotor), through measurement of **electrochemical skin conductance** and **sweat volume following electrical stimulation**
- Used to assess the degree of **neurologic damage and autonomic dysfunction** due to degeneration of **small nerve fibers**

QSART, quantitative sudomotor axon reflex test.
1.Shin & Robinson-Papp. *Mt Sinai J Med* 2012;79:733–48; 2. Illigens & Gibbons. *Clin Auton Res* 2009;19:79–87.

Autonomic testing[1,2]

Neuropathy assessments

- Includes measurement of **heart rate variability with deep breathing** to detect cardiovagal dysfunction in various autonomic disorders
  - Heart rate variability in response to deep breathing is notably depressed in patients with autonomic neuropathy

- **Tilt table test** monitors changes to blood pressure and heart rate in response to position changes
  - Aids in differentiating between forms of neurocardiogenic syncope, orthostatic hypotension, and non-cardiovascular conditions

1. Shields. *Cleve Clin J Med* 2009;76(Suppl 2): S37–40; 2. Teodorovich and Swissa. *World J Cardiol.* 2016;8(3):277–82.

Electrocardiogram (ECG)[1-3]

Cardiomyopathy assessments

- An ECG monitors the **electrical activity of the heart**. The components of the signal, referred to as “waves,” are classified as “P,” “Q,” “R,” “S,” or “T”
- Presence of **low QRS voltage, a pseudoinfarction pattern, atrioventricular block**, and **bundle branch block** can raise suspicion of **ATTR** ECG assessments are readily available tests that assist in raising the index of clinical suspicion

ATTR, transthyretin amyloidosis; ECG, electrocardiogram.

1. Ruberg & Berk. *Circulation.* 2012;126:1286–300; 2. Dharmarajan et al. *J Am Geriatr. Soc* 2012;60:765–74; 3. Gertz et al. *BMC Fam Pract.* 2020;21:198.

Echocardiogram[1-3]

Cardiomyopathy assessments

- 2D and 3D echocardiograms are used to **identify structural abnormalities** in the heart resulting from amyloid deposition
  - Patients often display ventricular wall thickening , atrial septal thickening, valve leaflets thickening, longitudinal strain impairment, increasing LV filling pressures, pericardial effusion, and increase speckling of the ventricular septum
  - 2D-speckle-tracking echo detection of diminished GLS with apical sparing may be indicative of ATTR[4,5]

ATTR, transthyretin amyloidosis; GLS, global longitudinal strain; LV, left ventricle.

1. Ruberg & Berk. *Circulation* 2012;126:1286–300; 2. Dharmarajan et al. *J Am Geriatr Soc* 2012;60:765–74; 3. Gertz et al. *BMC Fam Pract* 2020;21:198; 4. Castano et al. *Curr Cardiovasc Risk Rep* 2017;11:17; 5. Phelan et al. *Heart* 2012;98:1442–8.

Cardiac Magnetic Resonance Imaging (MRI)[1,2]

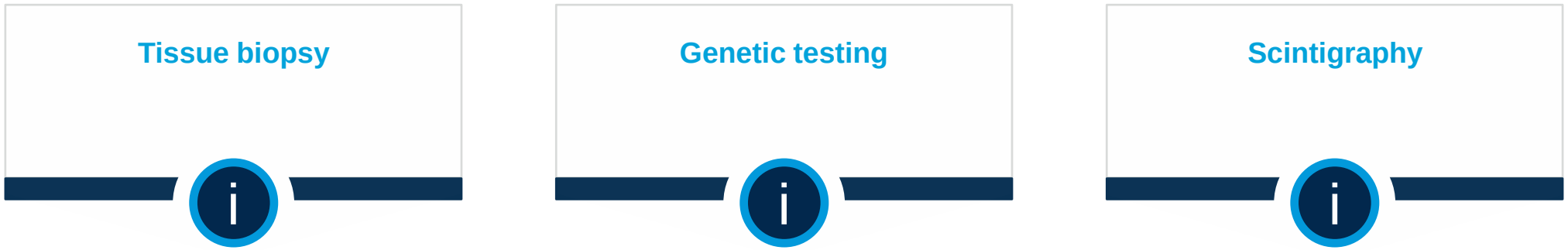
Cardiomyopathy assessments

- MRI can assess structural and functional characteristics of cardiac tissue affected by TTR deposition
  - Late gadolinium enhancement and T1 mapping can assess extracellular volume, interstitial expansion, and T1 relaxation
  - Patients often display biventricular wall thickening, increase in LV mass, diffuse subendocardial or transmural late gadolinium enhancement, increased native noncontrast T1 and extracellular volume
- MRI can raise suspicion of ATTR, but its use is limited by pacemakers and renal impairment

ATTR, transthyretin amyloidosis; LV, left ventricle; MRI, magnetic resonance imaging; TTR, transthyretin.

1. Gertz et al. *BMC Fam Pract* 2020;21:198; 2. Castano et al. *Curr Cardiovasc Risk Rep* 2017;11:17.

Confirmatory Diagnostic Tools for ATTR[1-3]



= Additional info

ATTR, transthyretin amyloidosis.

1. Ruberg & Berk. *Circulation.* 2012;126:1286–300; 2. Rapezzi et al. *Nat Rev Cardiol.* 2010;398–408; 3. Ando et al. *Orphanet J Rare Dis.* 2013;8:31.

Tissue biopsy[1-5]

- Biopsy of cardiac tissue **detects TTR amyloid deposition**, although SC fat aspirates of the abdominal wall, kidney, skin, gastric, or rectal mucosa can also be used
  - Amyloidosis of any type can be **detected based on Congo red staining**
  - Immunostaining of tissue with TTR antiserum can confirm diagnosis of ATTR
- Sensitivity may vary significantly by biopsy site (12%-41.7% [fat biopsy] - 99% [cardiac biopsy]); amyloid can be missing/infrequent in biopsy sample

ATTR, transthyretin amyloidosis; SC, subcutaneous; TTR, transthyretin.

1. Ruberg & Berk. *Circulation.* 2012;126:1286–300; 2. Rapezzi et al. *Nat Rev Cardiol.* 2010;398–408; 3. Nishi et al. *Circ J.* 2022;86:1113–20; 4. Garcia et al. *Hum Pathol.* 2018;72:71–9; 5. Hansen et al. *Molecules.* 2021;26:3649.

Genetic testing[1-3]

- Genetic testing can be performed to **detect and identify *TTR* variants** that will **distinguish** hATTR from wtATTR amyloidosis
- **PCR is used to detect *TTR* variants** (pathogenic and variants of unknown significance)
- Proteomic analysis of laser-dissected tissues using mass spectrometry has high specificity and sensitivity for identification of amyloid type and can distinguish between wild-type and variant TTR

hATTR, hereditary ATTR; PCR, polymerase chain reaction; TTR, transthyretin; wtATTR, wild-type ATTR.

1. Ando et al. *Orphanet J Rare Dis.* 2013;8:31; 2. Vrana et al. *Haematologica.* 2014;99:1239–47; 3. Tsuchiya et al. *Liver Transplant.* 2008;14:563–70.

Scintigraphy[1-5]

- Scintigraphy identifies amyloid buildup in certain parts of the body using radioactive tracers that **bind to amyloid deposits**
  - ^{99m}Tc-aprotinin, ^{123}I-MIBG, ^{123}I-SAP, ^{99m}Tc-PYP, and ^{99m}Tc-DPD all have use within ATTR
- **Specific/sensitive for diagnosis** after exclusion of plasma cell dyscrasia
  - Semiquantitative methods measuring H/CL and HWB ratios can aid definitive diagnosis of ATTR
  - Becoming most frequently used non-invasive diagnostic technique

ATTR, transthyretin amyloidosis;H/CL, heart-to-contralateral chest; HWB, heart/whole body; I-MIBG, iodine-131 meta-iodobenzylguanidine; I-SAP, iodine-123 labelled serum amyloid P component; Tc-DPD, technetium-3,3-diphosphono-1,2-pyrophosphate; Tc-PYP, technetium pyrophosphate; TTR, transthyretin; wtATTR, wild-type ATTR.

1. Ruberg & Berk. *Circulation.* 2012;126:1286–300; 2. Dharmarajan et al. *J Am Geriatr* Soc. 2012;60:765–74; 3. Castano et al. *Curr Cardiovasc Risk Rep.* 2017;11:17; 4. Gillmore et al. *Circulation.* 2016;133:2404–12; 5. Maurer et al. *Circ Heart Fail.* 2019;12:e006075

Summary

- ATTR is a multisystemic, rapidly progressive, debilitating, and fatal disease caused by misfolded TTR accumulating as amyloid deposits in multiple organs and tissues including nerves, heart, and GI tract [1-4]
  - Patients diagnosed with hATTR and wtATTR amyloidosis have a median survival of 4.7[5] and 2.5-5.5 years,[6-8] respectively
- ATTR remains underdiagnosed or misdiagnosed[4,9,10]
- Patients with ATTR experience substantial burden, including reduced QoL[11-14] and functional impairment[6,15]

There remains a need for health care professionals to:

1	2	3	4
Recognize the constellation of red-flag symptoms of ATTR[16,17]	**Collaborate with a multidisciplinary team for a potential diagnosis[16,17]**	**Employ the diagnostic algorithm and confirmatory diagnostic tools to verify diagnosis[17-19]**	**Assess progression of disease following treatment and provide patient with holistic care (mental, physical, and social support)[20,21]**

ATTR, transthyretin amyloidosis; hATTR, hereditary ATTR; wtATTR, wild-type ATTR; GI, gastrointestinal; QoL, quality of life; TTR, transthyretin.

1. Hanna. *Curr Heart Fail Rep.* 2014;11:50–7; 2. Mohty et al. *Arch Cardiovasc Dis.* 2013;106:528–40; 3. Adams et al. *Neurology.* 2015;85:675–82; 4. Maurer et al. *Circ Heart Fail.* 2019;12:e006075; 5. Swiecicki et al. *Amyloid.* 2015;22:123–31; 6. Lane et al. *Circulation.* 2019;140:16–26; 7. Aus dem Siepen et al. *Clin Res Cardiol.* 2018;107(2):158–69; 8. Givens et al. *Aging health.* 2013;9(2):229–35; 9. Hawkins et al. *Ann Med.* 2015;47:625–38; 10. Castano et al. *Heart Fail Rev.* 2015;20:163–78; 11. Coehlo et al. *Muscle Nerve.* 2017;55:323–32; 12. Vinik et al. *J Peripher Nerv Syst.* 2014;19:104–14; 13. Ines et al. *ISPOR Congress* 2015. Poster N21; 14. Obici et al. *Amyloid.* 2020;27:153–62; 15. Bolte et al. *Orphanet J Rare Dis* 2020;15:287; 16. Nativi-Nicolau et al. *Heart Fail Rev*. 2022;27(3):785–93; 17. Kittleson et al. *JACC.* 2023; 81(11):1076–176; 18. Namiranian and Geisler. *Am J Med.* 2022;135 Suppl 1:S13–19; 19. Ando et al. *Orphanet J Rare Dis.* 2013;8:31; 20. Adams et al. *Orphanet J Rare Dis.* 2021;16:411; 21. Obici et al. *BMJ Open.* 2023;13:e073130.

Alnylam®