Supplementary material

Szczygieł JA, Michałek P, Truszkowska G et al. Clinical features, etiology and survival in patients with restrictive cardiomyopathy: A single center experience. Kardiol Pol. 2023.

Table S1. Features of amyloidosis assessed during cardiac amyloidosis screening in all patients with restrictive cardiomyopathy (n = 36)

History	Cardiological symptoms and signs	Digestive system symptoms[a]
Age >30 yrs. No family history of cardiomyopathy Family history of AL amyloidosis, MM, MGUS Family history of ATTR amyloidosis Harmful working conditions, heavy metal fumes, varnishes Obesity in the past Chronic inflammation	≤12 mos. of HF symptoms Rapidly progressive HF Orthostatic hypotension Hypertension resolution Anti-hypertensive drugs intolerance Peripheral edema	≥10 kg weight loss Early satiety Loss of appetite Meat aversion Persistent diarrhea (due to milk dishes) Evening flatulence and stomach pains Persistent constipation Dysgeusia, xerostomia
Other symptoms and signs	**Physical examination**	**Laboratory abnormalities**
Carpal tunnel syndrome Biceps tendon rupture Spinal stenosis Nasal speech, hoarseness Symmetric painful neuropathy, numbness Urine retention or incontinence Frequent infections Urine foaming Bone pains, fractures	Macroglossia Submandibular swelling Shoulder pad Periorbital purpura Petechiae Pleural effusion Ascites Nail lesions Cachexia	Elevated hs-TnT and NT-proBNP Rapidly progressive kidney failure Nephrotic syndrome Anemia Pancytopenia Hypoalbuminemia Hyponatremia Hypercalcemia Increased activity of LDH, ALP or GGT
Electrocardiogram	**Echocardiography**	**Cardiovascular magnetic resonance**

Low amplitude of QRS complexes Pseudo-infarct pattern	≥ 12 mm LV wall thickening RV free wall thickening Thickening of valve leaflets Interatrial septum thickening Right atrium dilation Myocardial granular sparking Decreased tissue Doppler velocities Low-flow/low-gradient aortic stenosis Dilated inferior vena cava Pericardial effusion Impaired global longitudinal strain with relative apical sparing ('cherry on the top' pattern)[b]	Diffuse subendocardial or transmural LGE Myocardial nulling before the blood pool Increased T1 and T2 mapping[b]

[a]High probability of amyloid deposits in gastric and duodenal biopsy; [b]These features were only assessed in a few patients

Abbreviations: AL – light-chain; ALP – alkaline phosphatase; ATTR – transthyretin amyloidosis, GGT – gamma-glutamyl transferase; HF – heart failure; hs-TnT – high-sensitive troponin T; LGE – late gadolinium enhancement; LDH – lactate dehydrogenase; LV – left ventricular; MGUS – monoclonal gammopathy of undetermined significance; MM – multiple myeloma; NT-proBNP – N-terminal-proB-type natriuretic peptide; RV – right ventricular

Table S2. Genes included in genetic analysis of 17 patients with non-amyloid restrictive cardiomyopathy

Gene name according to the HGNC	Encoded protein
ABCC6	ATP-binding cassette subfamily C member 6
ACTC1[a]	Cardiac actin
ACTN2[a]	Alpha actinin 2
ALPK3	Alpha kinase 3
APOA1	Apolipoprotein A-1
ATAD3A	ATP-ase family AAA domain-containing 3A
BAG3[a]	Bcl2-associated athanogene 3
CACNA1C	Calcium voltage-gated channel subunit alpha 1C
CRYAB[a]	Alpha B crystallin
DCBLD2[a]	Discoidin cub and lccl domain-containing protein 2
DES[a]	Desmin
FHL1	Four and half LIM domains 1
FHOD3	Formin homology 2 domain-containing 3

FLNC[a]	Filamin C
GLA	Galactosidase alpha
GYG1	Glycogenin 1
HFE	Homeostatic iron regulator
JPH2	Junctophilin 2
LAMP2	Lysosomal associated membrane protein 2
LMNA[a]	Lamin A/C
MYBPC3[a]	Cardiac myosin-binding protein C
MYH7[a]	Beta myosin heavy chain
MYL2[a]	Cardiac regulatory myosin light chain
MYL3[a]	Essential myosin light chain 3
MYPN[a]	Myopalladin
PLN	Phospholamban
PRKAG2	Protein kinase AMP-activated non-catalytic subunit gamma 2
RPS6KB1	Ribosomal protein S6 kinase B1
TMEM87B[a]	Transmembrane protein 87 B
TNNC1[a]	Cardiac troponin C
TNNI3[a]	Cardiac troponin I
TNNT2[a]	Cardiac troponin T
TPM1[a]	Tropomyosin
TRIM63	Tripartite motif-containing 63
TTN[a]	Titin
TTR	Transthyretin

[a]Primary genetic restrictive cardiomyopathy

Abbreviations: HGNC – HUGO Gene Nomenclature Committee

Table S3. Clinical characteristics and genetic testing results of 8 patients with negative cardiac amyloidosis screening (biopsy not performed)

Patient no. in Table S4	Age (yrs.)	Sex	History duration (mos.)	Clinical details	Genetic testing[a] result	Survival (mos.)
1.	45	F	84	Symptoms of Anderson-Fabry disease	*GLA* p	1, OHT
4.	65	M	12	Systemic sclerosis	not performed	28, died

6.	20	F	84	Family history of cardiomyopathy	*MYH7* lp	69
8.	42	F	55	Scoliosis; CMR: LV IVS — 11 mm, PW — 8 mm, singular LGE area not typical for amyloidosis	*FLNC* lp *TTN* vus	10, died
9.	55	M	38	Family history of cardiomyopathy; CMR: LV IVS — 10 mm, LV PW — 10 mm, no LGE areas	*FLNC* vus	36, died
12.	44	M	63	Family history of cardiomyopathy; normal sFLC	*MYH7* lp	58
17.	18	F	60	Myopathic changes in muscle biopsy	*BAG3* p	50, died
18.	37	M	6	Family history of cardiomyopathy — relative of Patient 11	*MYBPC3* p	61

[a]Genetic testing included *TTR* gene analysis as mentioned in Table S2; in Patient 1 only *GLA* gene analysis by Sanger sequencing was performed

Abbreviations: CMR – cardiovascular magnetic resonance; F – female; IVS – interventricular septum; lp – likely pathogenic gene variant; M – male; OHT – orthotopic heart transplantation; p – pathogenic gene variant; PW – posterior wall; sFLC – serum free light chains, vus – gene variant of uncertain significance; others – see Tables S1 and S2

Table S4. Details about genetic testing in 18 patients with non-amyloid restrictive cardiomyopathy

No.	Age (yrs.)	Genetic testing method	Gene	Variant position (hg38), nucleotide and amino acid change	ACMG classification	ACMG criteria	Main references (PMID)
1.[a]	45	Commercial testing, SGS	*GLA*	chrX-101407766-G-T, NM_000169.3:c.138C>A (p.His46Gln)	Pathogenic	PM1, PM2, PM5, PP3	novel variant

			GLA	chrX-101407751-C-G, NM_000169.3:c.153G>C (p.Met51Ile)	Pathogenic	PS1, PM1, PM2, PP3	in late onset A-F disease 30477121
			GLA	chrX-101407737-C-A, NM_000169.3:c.167G>T (p.Cys56Phe)	Pathogenic	PS3, PM1, PM5, PP3, PM2	7531540, 25382311, 24386359
2.	27[b]	TSO	*TTN*[d]	chr2-178534401-A-G, NM_001267550.2:c.102214T>C (p.Trp34072Arg), rs375159973	Likely pathogenic	PS1, PM2, PP3, PP5	in patients with DCM or core myopathy with second truncating variant in TTN [24105469, 31983221, 32778822]

3.[a]	35[b]	WES	*MYH7*	chr14-23429005-G-A, NM_000257.4:c.1357C>T (p.Arg453Cys), rs121913625	Pathogenic	PS1, PS2, PM1, PM2, PP2, PP3, PP5	patient published by us [32013205], 33673806, 33586461, 31513939, 29907873
4.	65	N/A (systemic sclerosis – genetic testing not performed)					
5.[a]	57[b]	Commercial testing, SGS	*GLA*	chrX-101403846-G-A, NM_000169.3:c.334C>T (p.Arg112Cys)	Pathogenic	PS1, PM1, PM2,, PP2, PP3, PP5	1315715, 30477121
6.[a]	20[b]	TSO	*MYH7*	14:23418243-G-T, NM_000257.4:c.4136C>A (p.Ala1379Asp)	Likely pathogenic	PM1, PP2, PM2, PM5, PP3	27532257
7.[a]	33	TSO	*TNNI3*	19:55151904-A-C, NM_000363.5:c.563T>G (p.Val188Gly)	Likely pathogenic	PP2, PM2, PM5, PP3	novel variant

8.[a]	42	TSO	*FLNC*	7:128851562-T-G, NM_001458.5:c.5776T>G p.Tyr1926Asp	Likely pathogenic	PP3, PM2	novel variant
			TTN	2:178776534-C>T, NM_001267550.2:c.5330G>A (p.Cys1777Tyr)	VUS	PM2, PP3	novel variant
9.	55[b]	WES	*FLNC*	7:128849405-G>A, NM_001458.5:c.5026G>A (p.Gly1676Arg)	VUS	PP3, PM2, PP1	novel variant
10.[a]	49	TSO	*PRKAG2*	7:151576440-A>G, NM_016203.4:c.877T>C (p.Phe293Leu)	Likely pathogenic	PS1, PM2, PP3	29255176
			BAG3	10:119676965-G>A, NM_004281.4:c.1411G>A (p.Glu471Lys), rs778496291	VUS	PM2, PP3	in DCM patient 30442290
11.	63	TSO	*MYBPC3*	11:047332813-C>A, NM_000256.3:c.3490+1G>T, rs397516020	Pathogenic	PVS1, PM2, PP5	18957093, 28615295, 29121657
12.	44[b]	TSO	*MYH7*	14:023425363-A>T, NM_000257.4:c.2342T>A (p.Leu781Gln)	Likely pathogenic	PM1, PM2, PP2, PP3	novel variant

13.	63	TSO	*MYBPC3*	chr11-47341990 C-G, NM_000256.3:c.1790+1G>C	Pathogenic	PVS1, PM2, PP5	22857948
			ACTN2	chr1-236762528 G-C, NM_001103.4:c.2594G>C	VUS	PM2, BP6	novel variant
14.[a]	40	TSO	Nothing to report				
15.[a]	50	TSO	*TNNI3*	19:055151859-C-T, NM_000363.4:c.608G>A (p.Gly203Asp)	Likely pathogenic	PM1, PM2, PM5, PP2, PP3	novel variant
16.	52	TSO	*TNNI3*	19:055154073-A-G, NM_000363.5:c.506T>C (p.Leu169Pro)	Likely pathogenic	PM1, PM2, PP2, PP3	novel variant
17.[a]	18	TSO	*BAG3*	chr10-119672373 C-T, NM_004281.4:c.626C>T (p.Pro209Leu) rs121918312	Pathogenic	PM2, PM5, PP3, PP5	29338979, 25728519, 21361913, 32453099
18.	37[b,c]	SGS of variant	*MYBPC3*	11:047332813-C>A, NM_000256.3:c.3490+1G>T, rs397516020	Pathogenic	PVS1, PM2, PP5	22857948

		detected in Patient 11					

[a]Female; [b]Positive family history of cardiomyopathy; [c]Patient 18 is a relative of Patient 11; [d]Homozygous

Abbreviations: ACMG – American College of Medical Genetics and Genomics; A-F disease – Anderson-Fabry disease; DCM – dilated cardiomyopathy; hg38 – Genome Reference Consortium Human Build 38; N/A – not applicable – the test not performed; PMID – PubMed identifier; SGS – Sanger sequencing; TSO – TruSight One Sequencing Panel; VUS – gene variant of uncertain significance; WES – whole exome sequencing; others — see Tables S1 and S2

Table S5. Significant correlations of GDF15 and sST2 in the total cohort of 36 patients with restrictive cardiomyopathy

Variable	GDF15		sST2	
	Correlation coefficient	*P*-value	Correlation coefficient	*P*-value
Creatinine	0.402	0.02	–	N/A
eGFR	–0.533	0.001	–	N/A
GDF15	N/A	N/A	0.512	0.001
hs-TnT	0.357	0.03	–	N/A
IVC diameter	0.403	0.02	–	N/A
NT-proBNP	0.719	<0.001	0.591	<0.001
PASP	0.448	0.01	–	N/A
sST2	0.512	0.001	N/A	N/A
TAPSE	–0.419	0.01	–	N/A

Abbreviations: eGFR, estimated glomerular filtration rate; GDF15, growth differentiation factor-15; IVC, inferior vena cava, N/A, not applicable; PASP, pulmonary artery systolic pressure; sST2, soluble suppression of tumorigenicity 2; TAPSE, tricuspid annulus plane systolic excursion others — see Table S1