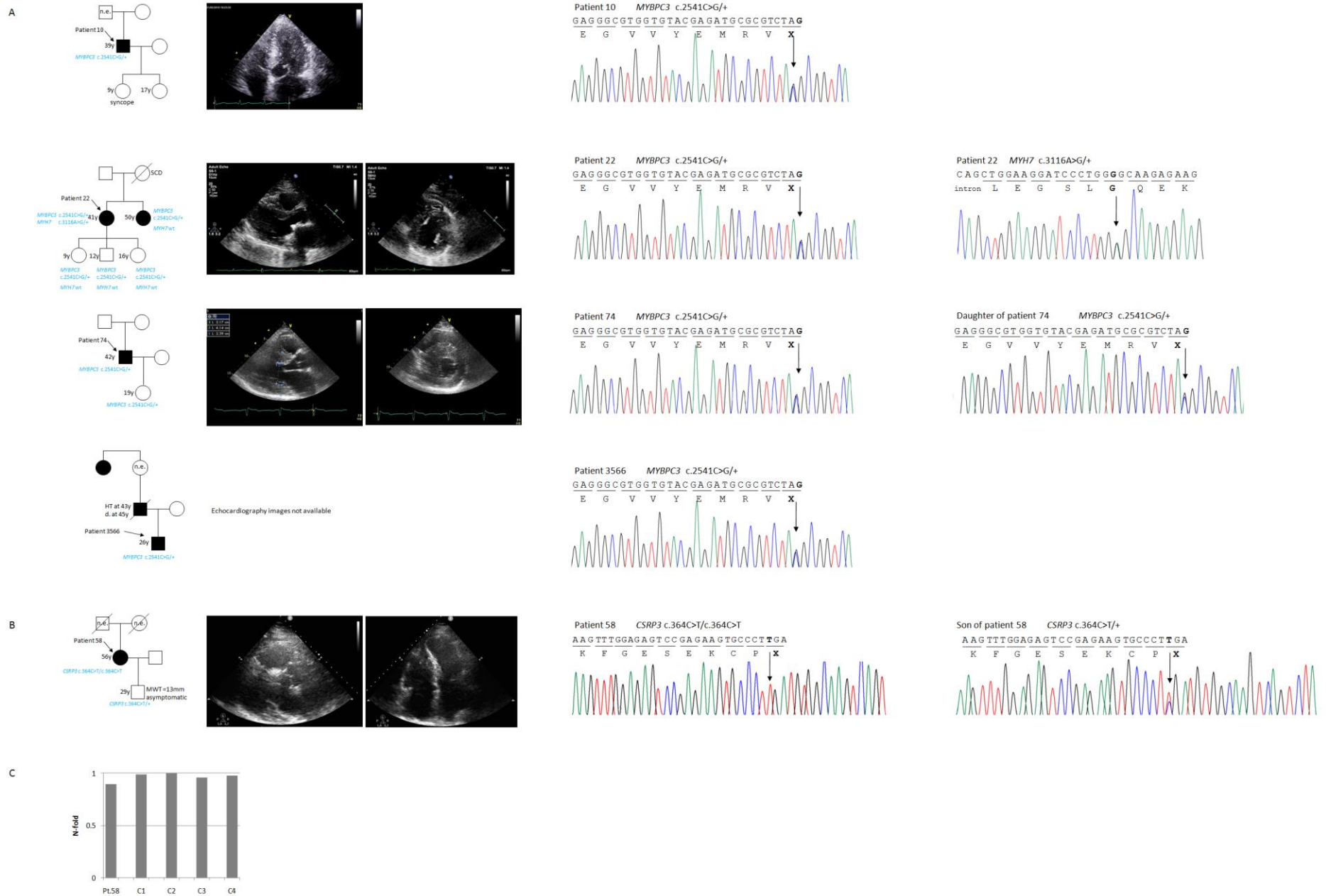


## **Supplementary material**

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*Lipari M, Wypasek E, Karpiński M, et al. Identification of a variant hotspot in MYBPC3 and of a novel CSRP3 autosomal recessive alteration in a cohort of Polish patients with hypertrophic cardiomyopathy. Pol Arch Intern Med. 2020; 130: 89-99. doi:10.20452/pamw.15130*

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**Figure S1.** Pedigrees, transthoracic echocardiography and Sanger sequencing chromatograms of cases carrying the (A) *MYBPC3* c.2541C>G variant and the (B) *CSRP3* c.364C>T null variant. Individuals with hypertrophic cardiomyopathy are displayed as black symbols. +: wild type allele; y: years at last clinical evaluation; d: age at death; n.e.: not evaluated; SCD: sudden cardiac death; HT: heart transplantation; MWT: maximal wall thickness; wt: wild type. Echocardiography images: (left) parasternal long axis view; (right) parasternal short axis view. (C). Graphical representation of the *CSRP3* qPCR results in case 58, showing the absence of copy number changes between nucleotide chr11:19207739 and chr11:19207865 encompassing the c.364C>T. The qPCR chart shows on the y-axis the N-fold changes in *CSRP3* copies normalized to *TERT*, relative to the copy number of the calibrator DNA (C2), according to the following equation: amount of target =  $2^{-\Delta\Delta Ct}$ . The samples included in the experiment are reported on the x-axis. C1, C2, C3 and C4 stand for four not affected controls.











XYLT1	2880	90.9	1137.89	1097	336.05	307	196.01	148	314.7	240	518.37	356	937.16	707	253.03	356.5	919.11	843	1097.71	1099	1080.7	1060.5	600.81	566	627.86	589	565.34	473	1185.45	1030.5	1145.29	1137	1044.17	1022.5	1028.83	946	119.04	111	235.32	240	173.08	113	230.52	228	1217.25	1167	646.61	579	282.69	214	157.08	146	90.07	35	283.15	283	374.2	362	818.61	726
YWHAE	768	96.5	649.04	355	144.85	103	161.95	129	212.76	199	340.67	49	732.37	138	162.41	176	429.99	262	573.49	261	574.37	340.5	246.35	191	206.61	178.5	223.64	164	411.66	248	599.56	319	498.65	232.5	499.04	317.5	59.6	33	114.96	92	95.84	31	115.4	117	608.16	297	269.02	207	120.98	138	98.59	94	25.13	10	131.69	110	195.97	186	332.83	235.5
YWHAG	744	100	1212.24	942	359.39	169	136.9	105	360.97	243	436.43	388	909.77	538	272.62	409	856.26	686	1070.58	789	1028.98	666	707.9	545	658.65	447	600.88	459	702.53	444	1117.47	740	969.56	639	952.17	654	129.28	124	264.33	249	162.28	125	257.6	230	1162.2	890	674.12	437	347.46	249	242.72	135	122.19	17	301.08	259	433.11	399	978.36	653
ZEB2	3645	100	814.55	702	251.25	208	239.75	188	296.55	286	380.09	300	811.14	296	288.5	270	626.68	582	758.33	727	782.06	693	492.35	378	463.3	332	412.45	334	576.11	553	849.23	746	700.35	643	771.61	682	86.44	72	164.26	153	117.41	78	178.04	160	833.07	766	470.16	388	235.95	179	133.32	113	75.87	44	199.4	181	327.44	283	591.97	453
ZFPM2	3456	100	841.34	705	255.1	205	159.88	137	259.49	183	343.74	188.5	768.17	169.5	275.87	284	645.49	538	730.27	603	730.51	653	433.15	370	428.55	353.5	367.95	317	552.18	460	763.25	627	657.55	544	666.36	646	80.81	69	168.87	136	109.54	80	167.48	155	864.8	750	444.74	394.5	236.57	153	124.09	97	71.46	30	190.21	157	313.62	235	552.43	461
ZIC3	1404	100	470.21	373	152.13	131	132.75	59	103.86	97	214.63	93	729.88	392	239.51	231	350.88	272	793.39	655	403.94	328	233.75	225	487.13	494	222.37	194	299.81	218	442.64	376	348.1	302	356.25	310	41.04	30	85.12	75	99.03	46	151.09	136	912.21	707	515.87	442	96.38	96	49.23	33	30.73	32	90.28	81	257.51	214	329.47	289
ZMPSTE24	1428	100	311.19	180	71.54	49	152.38	117	145.47	174	163.2	39	470.38	362	86.05	68	232.02	138	297.88	179	298.91	203	138.36	115	116.77	109	114.36	110	194.91	124	288.26	154	247.99	154	250.25	169	33.75	25	57.18	41	47.57	17	61.61	57	291.33	172	149.59	113	62.3	53	45.14	39	16.39	7	71.95	56	116.94	75	143.57	135

M: mean; Mdm: Median; nt: nucleotide; Pt: patient



**Table S2.** PCR primers used for the Sanger validation of the identified variants.

Gene	Variant	Primers	
<i>CSRP3</i>	c.364C>T	F	5'-tttctctcattcctccaaa-3'
		R	5'-ctctttgccaagggaaatc-3'
<i>GUSB</i>	c.454G>A	F	5'-gtcagtcaggtattggatgg-3'
		R	5'-tagcctgatctacccatt-3'
<i>GYG1</i>	c.304G>C	F	5'-ttctagtaattgatggagaagg-3'
		R	5'-agtggttctatctgaactttacc-3'
<i>MYBPC3</i>	c.772G>A	F	5'-gccactcccagttctcttta-3'
		R	5'-ggcatcctccttagtgttgg-3'
<i>MYBPC3</i>	c.821+5G>A	F	5'-catgaatgggcaagtctgtg-3'
		R	5'-aatcatccccagccctga-3'
<i>MYBPC3</i>	c.1483C>T	F	5'-cgggtggagtttgagtgtg-3'
		R	5'-ctcccctacagggttaggtg-3'
<i>MYBPC3</i>	c.1513A>T	F	5'-cgggtggagtttgagtgtg-3'
		R	5'-ctcccctacagggttaggtg-3'
<i>MYBPC3</i>	c.2541C>G	F	5'-caaacagatccgaggggaagg-3'
		R	5'-gtatccccagtcgaggatga-3'
<i>MYH7</i>	c.428G>A	F	5'-ttctctaactccaaaatcacca-3'
		R	5'-ggggaaagaggctgagtcta-3'
<i>MYH7</i>	c.3116A>G	F	5'-ggtttccaagtctgaaca-3'
		R	5'-gctgtcttgggtctgcttgt-3'
<i>MYH7</i>	c.4190T>G	F	5'-ctgtcctcaggcttctccag-3'
		R	5'-ctctggcctctcactgaacc-3'
<i>PMM2</i>	c.691G>A	F	5'-ggagcctcatctgtactt-3'
		R	5'-ctaataccaccctgatacg-3'
<i>SCO2</i>	c.418G>A	F	5'-tggaagtctggacgtag-3'
		R	5'-gcttttgcaggcagg-3'
<i>TNNT2</i>	c.304C>T	F	5'-ctagcccaccatctctcc-3'
		R	5'-cgaggaaggctgtctgga-3'

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