

Seno-destructive smooth muscle cells in the ascending aorta of patients with bicuspid aortic valve disease

Supplementary Materials and Methods

Table S1 Primer sequences for reverse transcription-quantitative PCR

Gene	Forward sequence	Reverse sequence
CDKN2A (P16 ^{Ink4a})	CTCGTGCTGATGCTACTGAGGA	GGTCGGCGCAGTTGGGCTCC
CDKN1A (P21)	AGGTGGACCTGGAGACTCTCAG	TCCTCTTGGAGAAGATCAGCCG
MMP1	ATGAAGCAGCCCAGATGTGGAG	TGGTCCACATCTGCTCTTGGCA
MMP2	AGCGAGTGGATGCCGCCTTAA	CATTCCAGGCATCTGCGATGAG
MMP3	CACTCACAGACCTGACTCGGTT	AAGCAGGATCACAGTTGGCTGG
MMP7	TCGGAGGAGATGCTCACTTCGA	GGATCAGAGGAATGTCCCATAACC
MMP8	CAACCTACTGGACCAAGCACAC	TGTAGCTGAGGATGCCTTCTCC
MMP9	GCCACTACTGTGCCTTTGAGTC	CCCTCAGAGAATCGCCAGTACT
MMP10	TCCAGGCTGTATGAAGGAGAGG	GGTAGGCATGAGCCAAACTGTG
MMP11	GAGAAGACGGACCTCACCTACA	CTCAGTAAAGGTGAGTGGCGTC
MMP12	GATGCTGTCACTACCGTGGGAA	CAATGCCAGATGGCAAGGTTGG
MMP13	CCTTGATGCCATTACCAGTCTCC	AAACAGCTCCGCATCAACCTGC
MMP14	CCTTGGACTGTCAGGAATGAGG	TTCTCCGTGTCATCCACTGGT
MMP15	CTGGCTCTTTCGAGAAGCGAAC	TCTCCTCGTTGAAGCGCCAGTA
MMP16	GATTCAGCCATTTGGTGGGAGG	CCCTTCCAGACTGTGATTGGC
TIMP1	GGAGAGTGTCTGCGGATACTTC	GCAGGTAGTGATGTGCAAGAGTC
TIMP2	ACCCTCTGTGACTTCATCGTGC	GGAGATGTAGCACGGGATCATG
TIMP3	TACCGAGGCTTCACCAAGATGC	CATCTTGCCATCATAGACGCGAC
TIMP4	CACTACCATCTGAACTGTGGCTG	GCTTTCGTTCCAACAGCCAGTC
COL1A1	GATTCCCTGGACCTAAAGGTGC	AGCCTCTCCATCTTTGCCAGCA
COL1A2	CCTGGTGCTAAAGGAGAAAAGAGG	ATCACCACGACTTCCAGCAGGA
COL3A1	TGGTCTGCAAGGAATGCCTGGA	TCTTCCCTGGGACACCATCAG
COL7A1	GTTGGAGAGAAAGGTGACGAGG	TGGTCTCCCTTTTCACCACAG
COL8A1	AGGAAGCCGTACCCAAGAAAGG	GGTATCCCATGACCTGGCAAAC
COL12A1	CAGTGCTGTAGTCAGCCTGAA	GGTCTTGTGGCTCTGTGTCCT
COL14A1	CACAAACCTCCTCAGCGGAATG	GGCTTGGAGATTGGTAACACCC
COL15A1	GGTGACACTGGTTTACCTGGCT	GCCTTCCAGAGGAATGTCTC
COL16A1	AACAGTGAGGGAGATCCTGGCT	CAACAGCACCAGGAAAACCTGG
CCL2 (MCP-1)	AGAATCACCAGCAGCAAGTGTCC	TCCTGAACCCACTTCTGCTTGG
CCL3 (MIP-1 α)	ACTTTGAGACGAGCAGCCAGTG	TTCTGGACCCACTCCTCACTG
IL1B	CCACAGACCTTCCAGGAGAATG	GTGCAGTTCAGTGATCGTACAGG
IL6	AGACAGCCACTCACCTCTTCAG	TTCTGCCAGTGCCTCTTTGCTG
TGFB1	TACCTGAACCCGTGTTGCTCTC	GTTGCTGAGGTATCGCCAGGAA
TNF (TNF- α)	CTCTTCTGCCTGCTGCACCTTG	ATGGGCTACAGGCTTGTCACTC
RNA18S5 (18S)	ACCCGTTGAACCCCATTCGTGA	GCCTCACTAAACCATCCAATCGG

Supplementary Tables

Table S2 Clinical information for subjects with TAV and non-aneurysmal ascending aorta

Patient	Surgery	Age	Sex	Ascending Aorta Diameter (cm)	Normalized Ascending Aorta Diameter (cm/m ²)
1	CABG	83	M	3.5	1.9
2	CABG	53	M	3.8	1.8
3	CABG	85	F	2.8	1.5
4	HT R	70	F	2.5	1.8
5	HT R	41	F	3.5	1.8
6	HT D	ND	ND	ND	ND
7	HT R	ND	ND	ND	ND
8	CABG	78	M	3.2	1.7
9	CABG	84	F	2.8	1.9
10	CABG	86	F	3.3	2.4
11	HT R	59	M	3.7	2.3
12	HT D	ND	ND	ND	ND
13	HT R	18	M	3.3	1.7
14	HT D	ND	ND	ND	ND
15	HT D	ND	ND	ND	ND

CABG, coronary artery bypass graft; HT R, heart transplant recipient; HT D heart transplant donor. ND, Not determined;

Table S3 Clinical information for patients with BAV and non-aneurysmal ascending aorta

Patient	Age	Sex	Aortic Valve Configuration Sievers Classification	Aortic Valve Dysfunction	Ascending Aorta Diameter (cm)	Normalized Ascending Aorta Diameter (cm/m²)
1	59	M	Type 1; R-L	AS	4.2	2.0
2	56	M	Type 1; R-L	AR	4.2	1.8
3	32	M	Type 1; R-L	AR	3.1	1.5
4	50	F	Type 1; R-L	AS	4.1	1.9
5	53	M	Type 1; R-L	AS	3.3	1.7
6	50	M	Type 0; R-L	AR	3.5	1.8
7	55	M	Type 1; R-L	AR	3.5	1.9
8	30	M	Type 1; R-L	AR	3.4	1.9
9	59	M	Type 0; Lateral	AS	4.3	2.0
10	22	F	Type 0; Lateral	AS	3.4	2.6

AR, aortic regurgitation (\geq moderate); AS, aortic stenosis (\geq moderate)

Table S4 Clinical information for patients with TAV-associated ascending aortic aneurysm

Patient	Age	Sex	Aortic Valve Dysfunction	Ascending Aorta Diameter (cm)	Normalized Ascending Aorta Diameter (cm/m²)
1	72	M	Mild AS	5.0	2.4
2	80	F	None	5.1	2.9
3	75	F	None	6.3	3.7
4	80	M	None	4.9	2.6
5	62	M	AR	5.6	2.8
6	81	F	AR	4.3	2.3
7	79	F	AR	6.5	3.3
8	69	F	None	4.9	2.7
9	74	M	Mild AR	6.8	3.4
10	33	M	Mild AR	5.3	2.5
11	55	F	None	4.5	2.5
12	69	M	None	5.3	2.7
13	70	M	Mild AR	5.8	2.5
14	64	M	AR	4.5	2.1

AR, aortic regurgitation (\geq moderate unless specified); AS, aortic stenosis (\geq moderate unless specified)

Table S5 Clinical information for patients with BAV-associated ascending aortic aneurysm

Patient	Age	Sex	Aortic Valve Configuration Sievers Classification	Aortic Valve Dysfunction	Ascending Aorta Diameter (cm)	Normalized Ascending Aorta Diameter (cm/m ²)
1	75	F	Type 1; R-L	AS	4.4	2.9
2	62	M	Type 1; R-N	AS	5.1	2.6
3	56	M	Type 1; R-L	AS	4.6	2.1
4	64	M	Type 1; R-N	AR/AS	5.4	2.7
5	55	M	Type 0; Lateral	AS	5.5	2.6
6	37	M	Type 1; R-N	None	5.1	2.0
7	31	M	Type 2; R-L, R-N	AR/AS	5.8	2.9
8	63	F	Type 1; R-L	AR	5.2	2.6
9	44	M	Type 1; R-L	AR	5.4	2.8
10	70	M	Type 1; R-L	AR	5.4	2.7
11	56	M	Type 1; R-L	AR	5.1	2.6
12	73	M	Type 1; R-L	AR	5.5	2.6
13	40	M	Type 1; R-L	AR	5.7	2.7
14	64	M	Type 1; R-L	AS	4.1	2.2
15	63	M	Type 1; R-L	AS	4.3	2.5
16	79	M	Type 1; R-L	AR	4.6	2.4
17	52	M	Type 1; R-L	AR/AS	6.1	2.7
18	55	M	Type 1; R-L	AR	5.3	2.4
19	74	M	Type 1; R-N	AR	4.4	2.2
20	56	M	Type 0; Lateral	AS	5.1	2.7
21	74	F	Type 1; R-N	AS	4.6	2.7
22	52	M	Type 1; R-L	AR	5.2	2.4
23	68	M	Type 1; R-L	AR/AS	5.9	3.0
24	62	F	Type 1; R-L	AS	5.4	2.7
25	43	F	Type 1; R-L	AS	5.3	2.9
26	75	M	Type 1; R-L	AS	5.4	2.6
27	77	M	Type 1; R-L	AR	4.8	2.5
28	21	M	Type 0; R-L	AR	4.4	2.2
29	59	M	Type 1; R-L	AR/AS	5.3	2.7

AR, aortic regurgitation (\geq moderate); AS, aortic stenosis (\geq moderate)