Fig. 1A

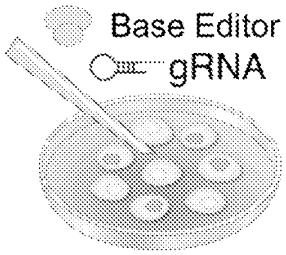


Fig. 1B

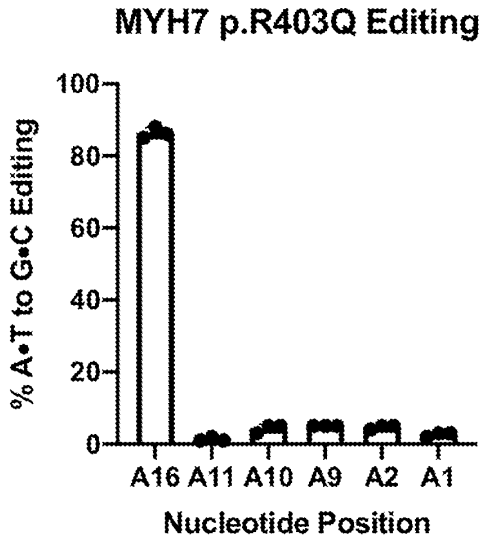


Fig. 1C

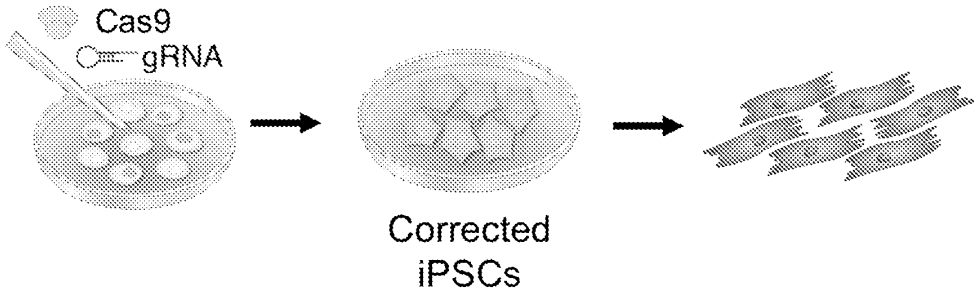

Cas9
gRNA
Corrected
iPSCs

Fig. 2A

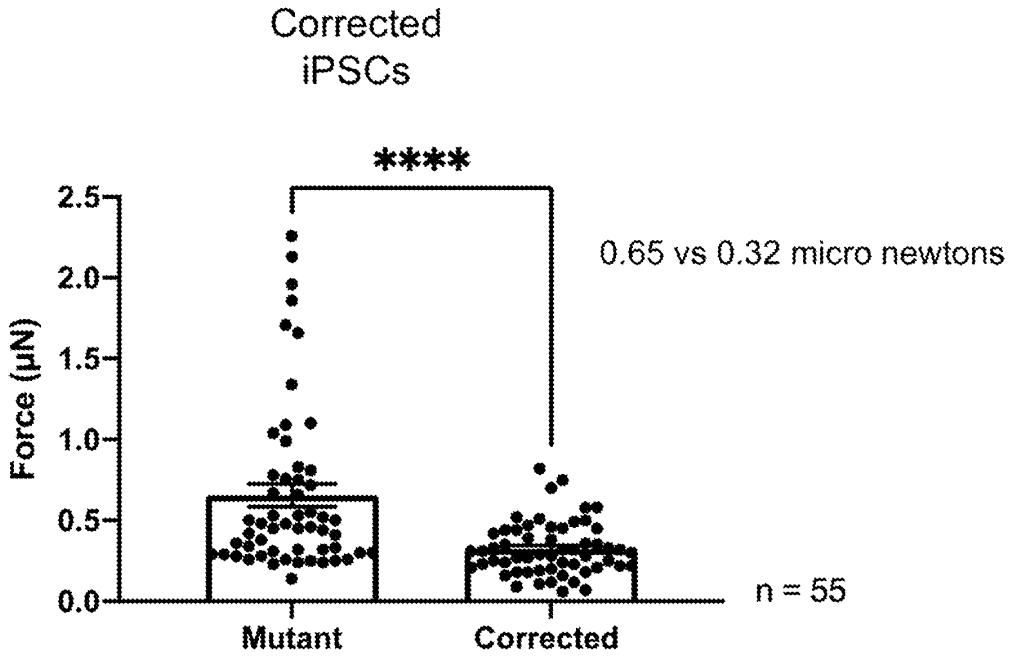

Corrected
iPSCs
****
0.65 vs 0.32 micro newtons
Force (µN)
2.5
2.0
1.5
1.0
0.5
0.0
n = 55
Mutant
Corrected

Fig. 2B

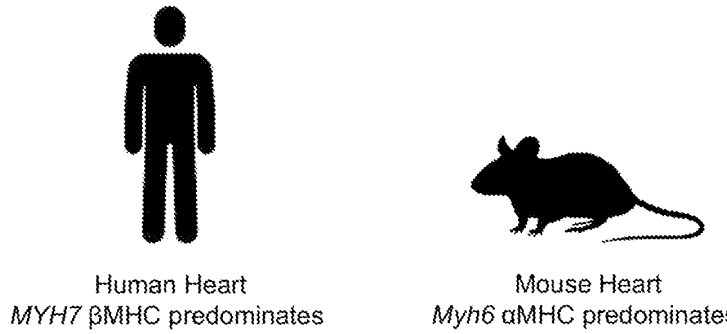

Human Heart
MYH7 βMHC predominates
Mouse Heart
Myh6 αMHC predominates

Fig. 3A

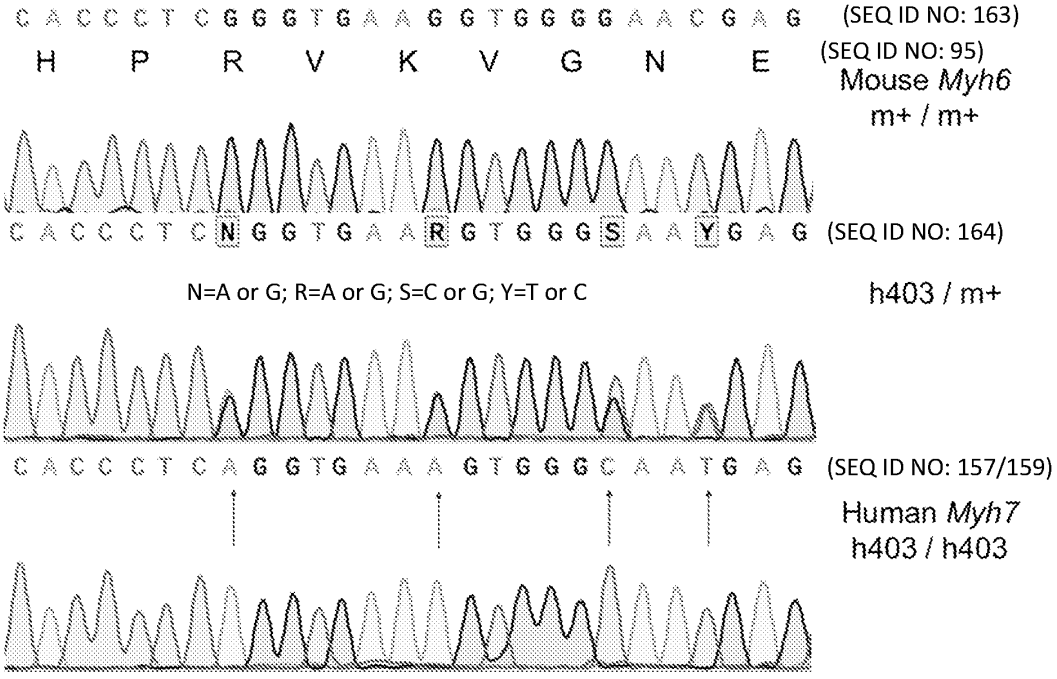

C A C C C T C G G G T G A A G G T G G G G A A C G A G (SEQ ID NO: 163)
H P R V K V G N E (SEQ ID NO: 95)
Mouse *Myh6*
m+ / m+
C A C C C T C N G G T G A A R G T G G G S A A Y G A G (SEQ ID NO: 164)
N=A or G; R=A or G; S=C or G; Y=T or C
h403 / m+
C A C C C T C A G G T G A A A G T G G G C A A T G A G (SEQ ID NO: 157/159)
Human *Myh7*
h403 / h403

Fig. 3B

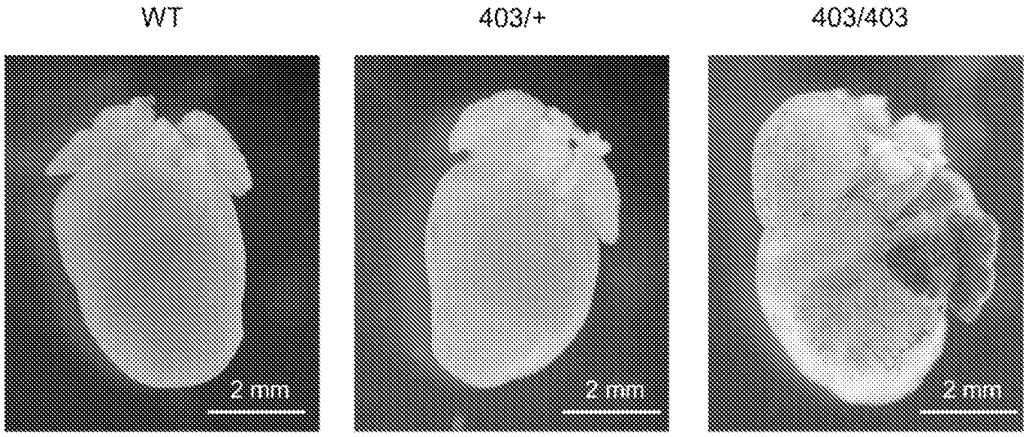


Fig. 4A Fig. 4B Fig. 4C

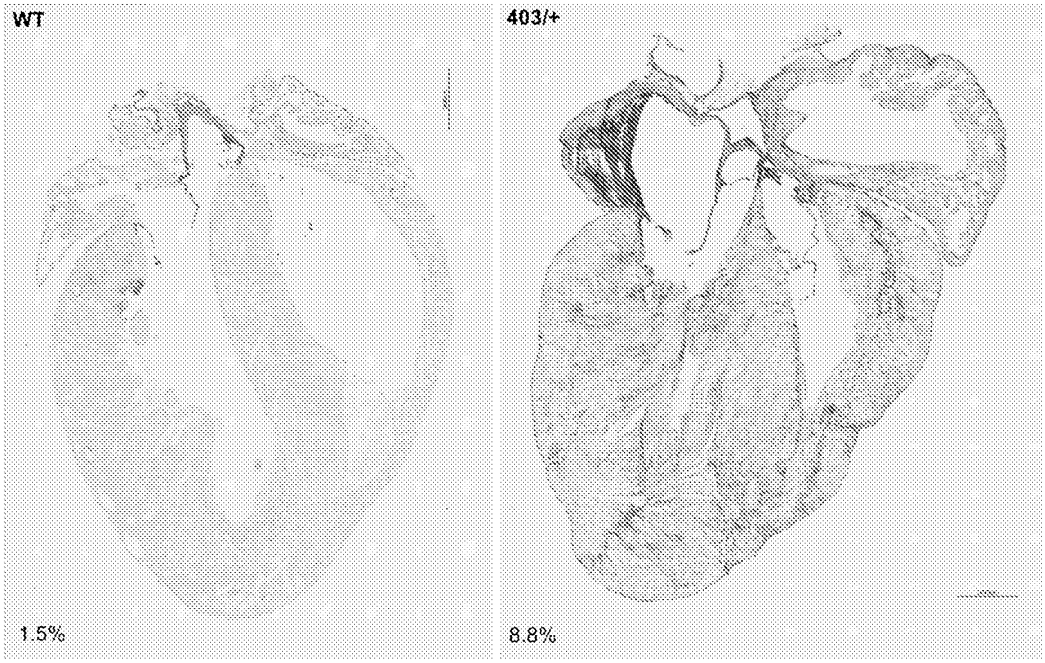
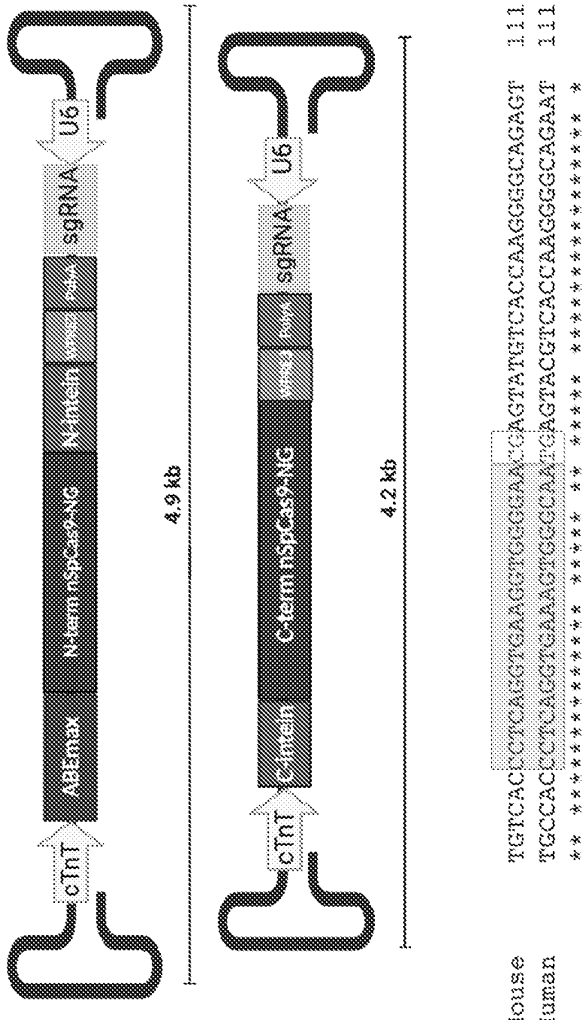


Fig. 4D Fig. 4E



Mouse TGTCACCCTCAAGGTGAAGGTCGGAACTCAGTATGTCACCAAGGGGCAGAGT 111 SEQ ID NO: 3
Human TGCCACCCTCACGTGAAAGTGGGCAATTCAGTACGTCACCAAGGGGCAGAAT 111 SEQ ID NO: 4
** ********** ***** ** ***** ********** * *

Mouse vs human sgRNAs for A-base editing at R403Q codon

Fig. 5

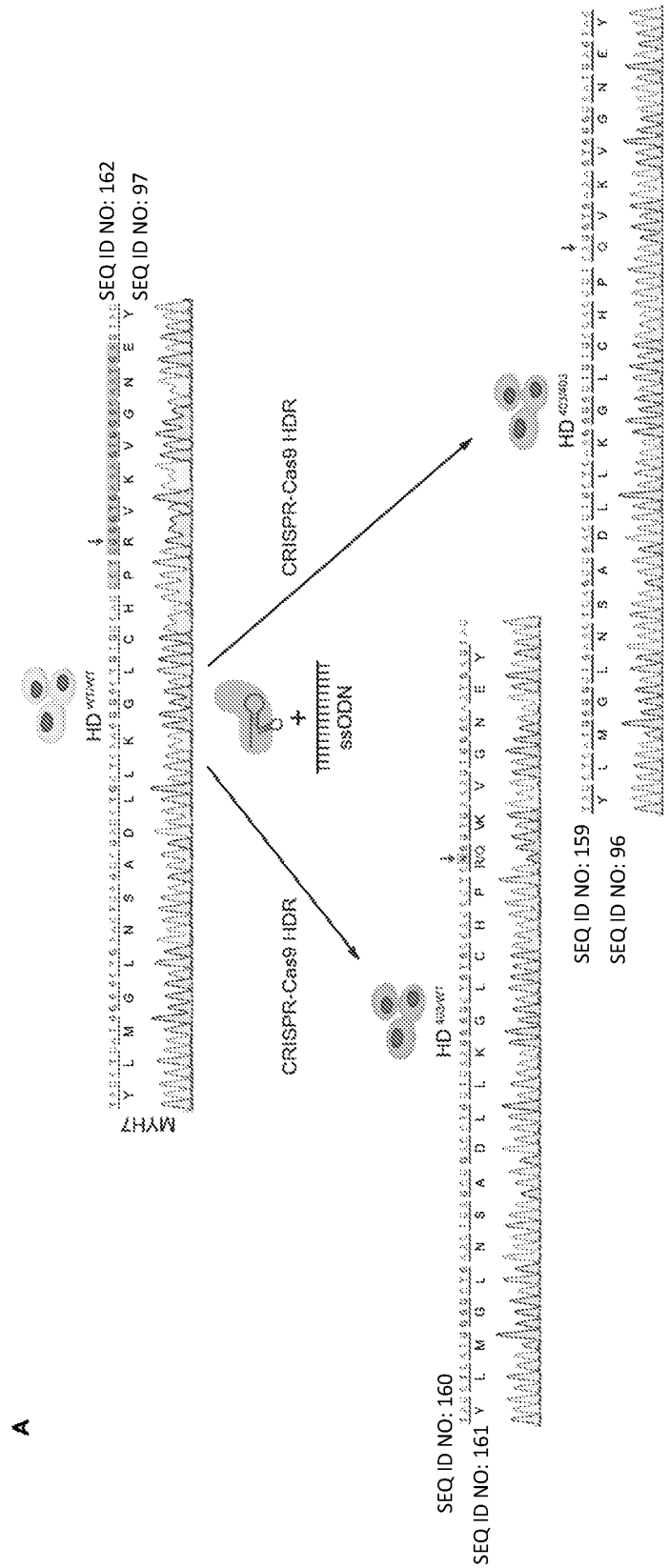

A
MYH7
HD WT/WT
SEQ ID NO: 162
SEQ ID NO: 97
CRISPR-Cas9 HDR
+
ssODN
CRISPR-Cas9 HDR
HD 403/403
HD 403/WT
SEQ ID NO: 160
SEQ ID NO: 161
SEQ ID NO: 159
SEQ ID NO: 96

Fig. 6A

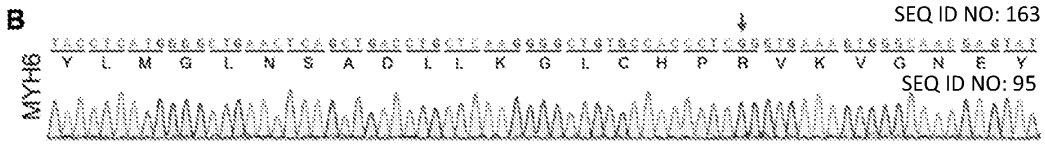

B
SEQ ID NO: 163
MYH6
Y L M G L N S A D L L K G L C H P R V K V G N E Y
SEQ ID NO: 95

Fig. 6B

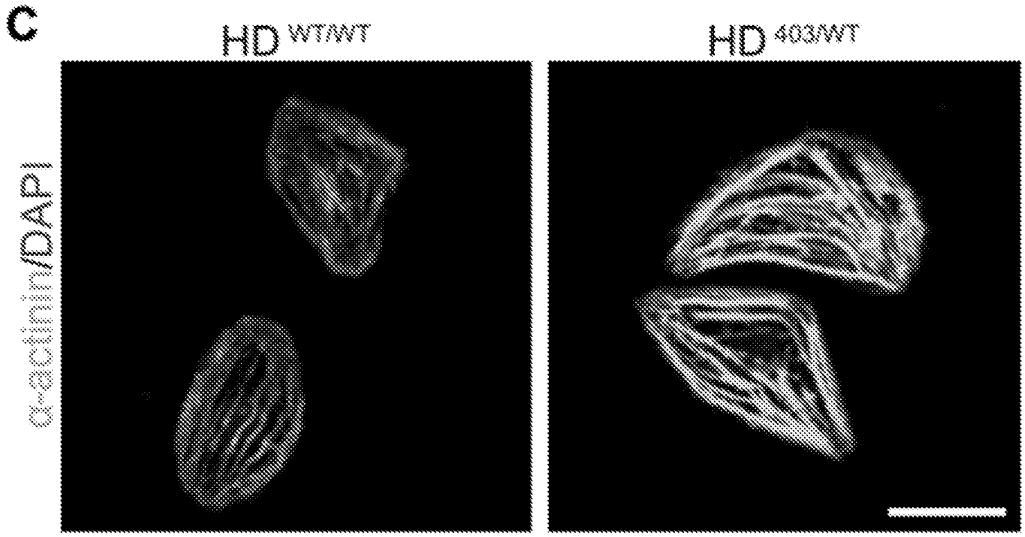

C
HD WT/WT
HD 403/WT
α-actinin/DAPI

Fig. 6C

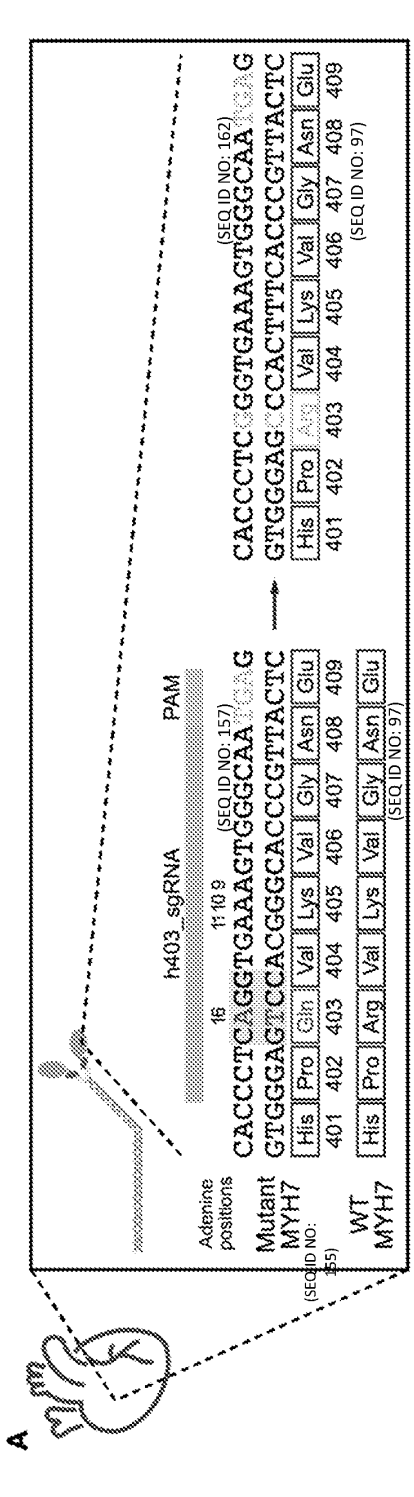

A
h403_sgRNA
PAM
16
11 10 9
Adenine positions
Mutant MYH7 (SEQ ID NO: 155)
CACCCTCAGGTGAAAGTGGGCAA (SEQ ID NO: 157)
GTGGGAGTCCACGGGCACCCGTTACTC
His Pro Gln Val Lys Val Gly Asn Glu
401 402 403 404 405 406 407 408 409
WT MYH7
His Pro Arg Val Lys Val Gly Asn Glu
(SEQ ID NO: 97)
CACCCTCGGTGAAAGTGGGCAA (SEQ ID NO: 162)
GTGGGAGCCACTTTCACCCGTTACTC
His Pro Arg Val Lys Val Gly Asn Glu
401 402 403 404 405 406 407 408 409
(SEQ ID NO: 97)

Fig. 7A

ABEmax
ABE8e
HD403/403
Cas9-SpCas9
Cas9-NG
Cas9-NG
Cas9-NG
Cas9-NG
Cas9-NG
+
h403_sgRNA

Fig. 7B

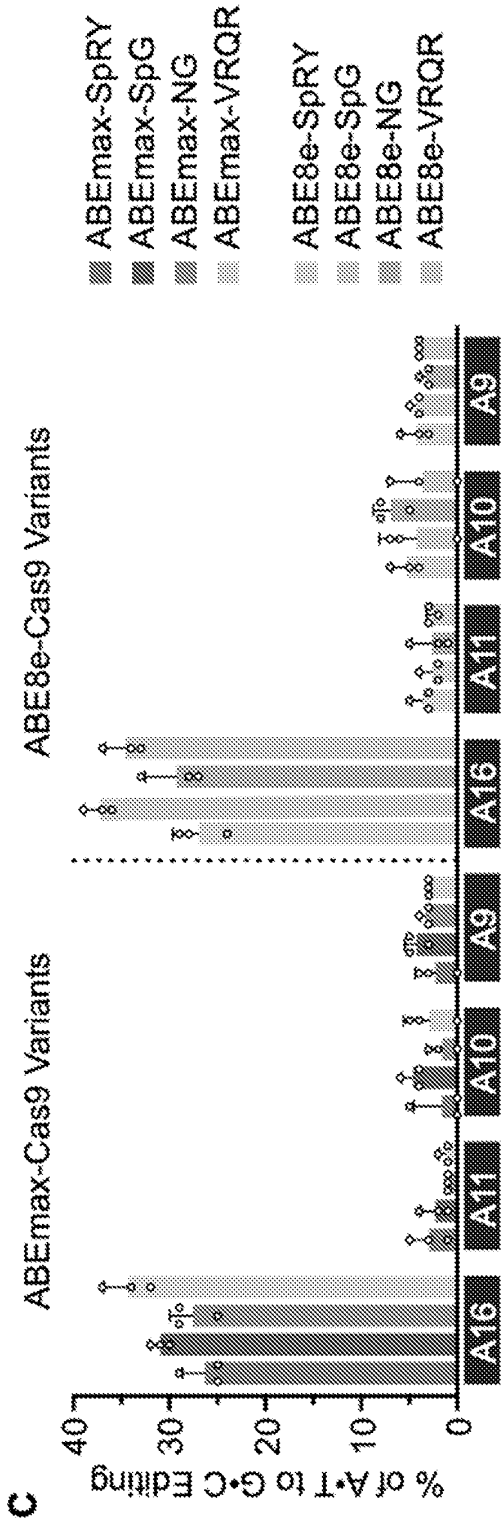

c
% of A•T to G•C Editing
0
10
20
30
40
ABEmax-Cas9 Variants
A16
A11
A10
A9
ABE8e-Cas9 Variants
A16
A11
A10
A9
ABEmax-SpRY
ABEmax-SpG
ABEmax-NG
ABEmax-VRQR
ABE8e-SpRY
ABE8e-SpG
ABE8e-NG
ABE8e-VRQR

Fig. 7C

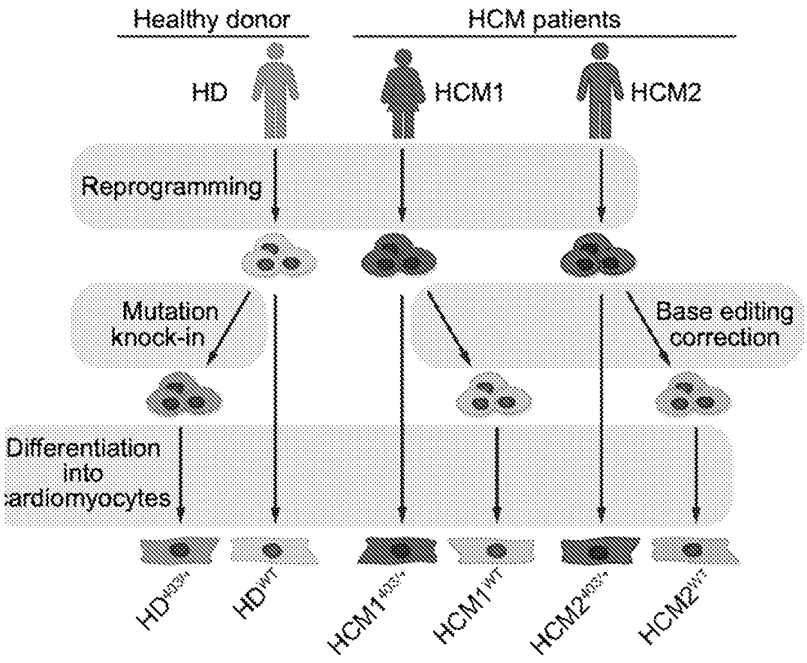

Healthy donor
HCM patients
HD
HCM1
HCM2
Reprogramming
Mutation knock-in
Base editing correction
Differentiation into cardiomyocytes
HD403/+
HD WT
HCM1 403/+
HCM1 WT
HCM2 403/+
HCM2 WT

Fig. 8A

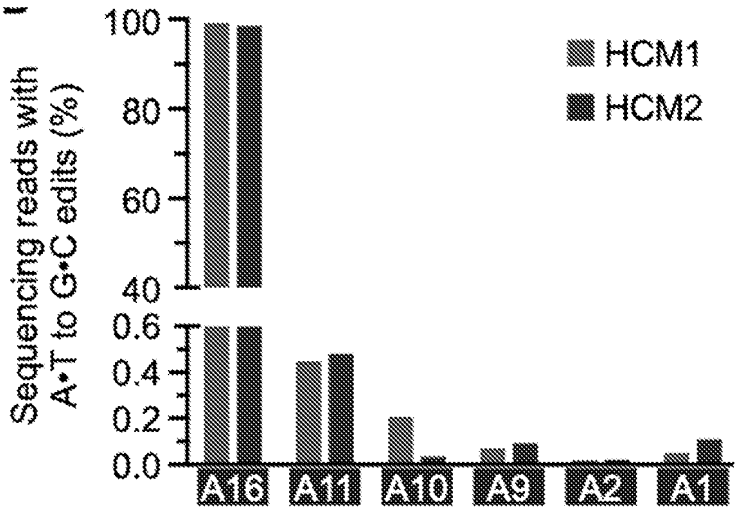

Sequencing reads with A•T to G•C edits (%)
100
80
60
40
0.6
0.4
0.2
0.0
HCM1
HCM2
A16
A11
A10
A9
A2
A1

Fig. 8B

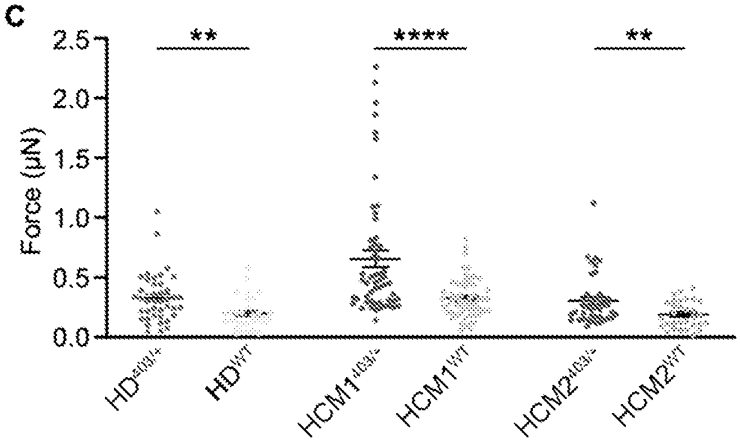

C
Force (μN)
2.5
2.0
1.5
1.0
0.5
0.0
**
****
**
HD403/+
HDWT
HCM1403/+
HCM1WT
HCM2403/+
HCM2WT

Fig. 8C

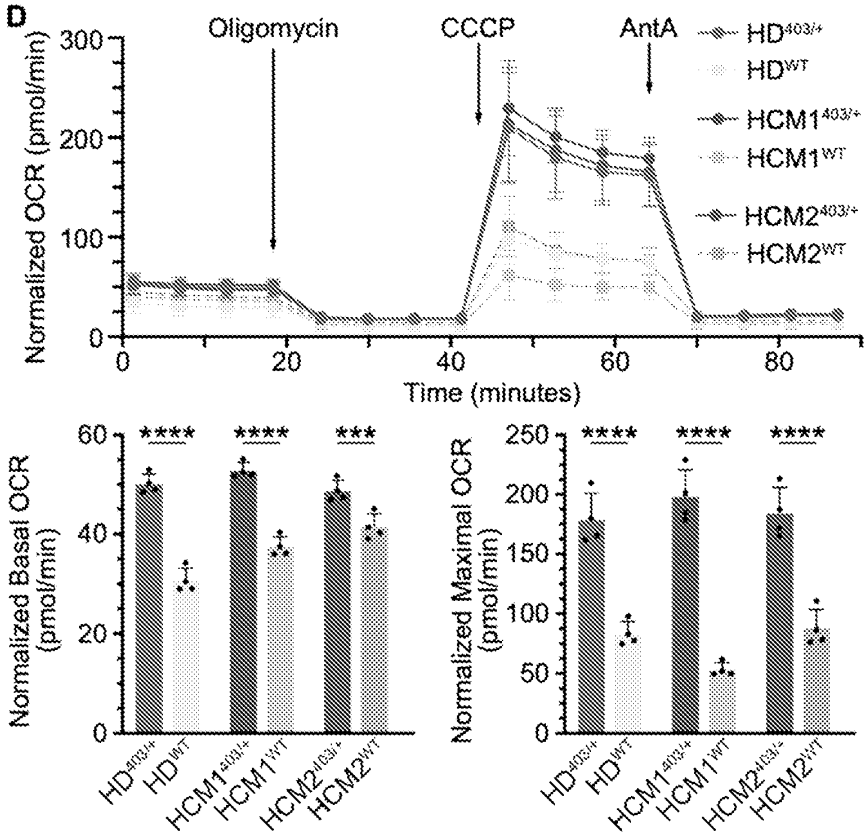

D
Normalized OCR (pmol/min)
300
200
100
0
Oligomycin
CCCP
AntA
HD403/+
HDWT
HCM1403/+
HCM1WT
HCM2403/+
HCM2WT
0
20
40
60
80
Time (minutes)
Normalized Basal OCR (pmol/min)
60
40
20
0
****
****
***
Normalized Maximal OCR (pmol/min)
250
200
150
100
50
0
****
****
****

Fig. 8D

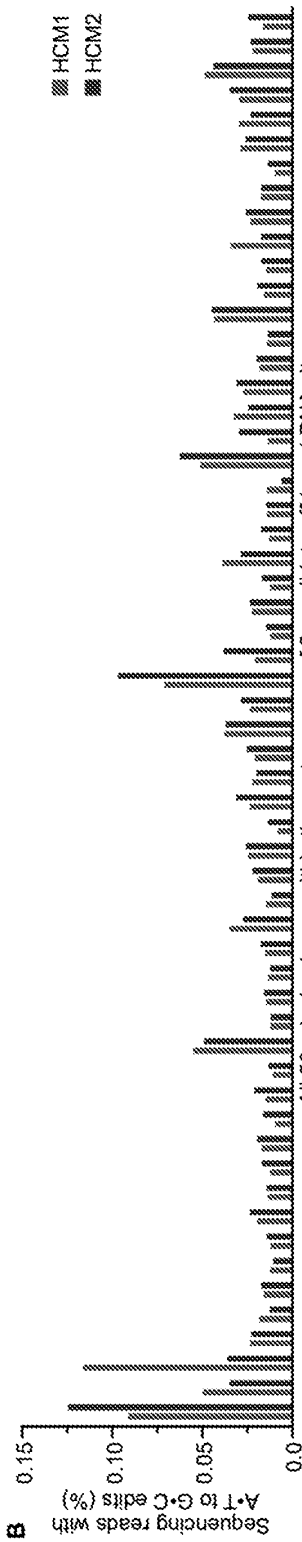

B
Sequencing reads with A•T to G•C edits (%)
0.15
0.10
0.05
0.0
HCM1
HCM2
All 58 adenine bases within the protospacers of 8 candidate off-target DNA sites

Fig. 9

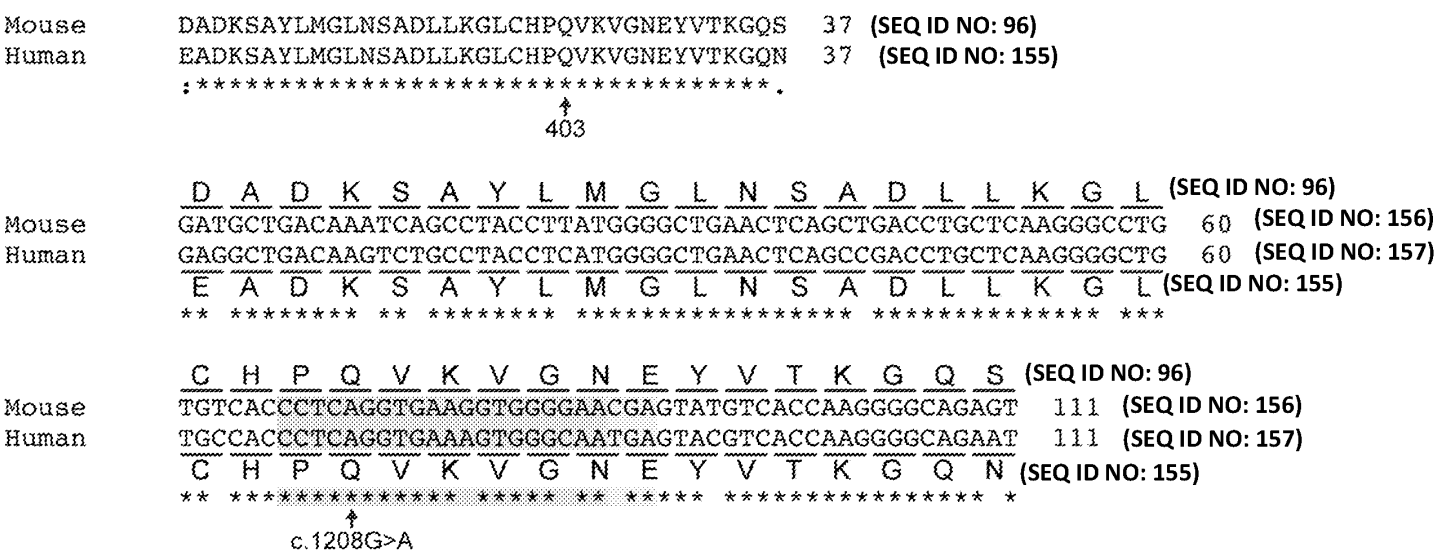

Mouse DADKSAYLMGLNSADLLKGLCHPQVKVGNEYVTKGQS 37 (SEQ ID NO: 96)
Human EADKSAYLMGLNSADLLKGLCHPQVKVGNEYVTKGQN 37 (SEQ ID NO: 155)
403
D A D K S A Y L M G L N S A D L L K G L (SEQ ID NO: 96)
Mouse GATGCTGACAAATCAGCCTACCTTATGGGGCTGAACTCAGCTGACCTGCTCAAGGGCCTG 60 (SEQ ID NO: 156)
Human GAGGCTGACAAGTCTGCCTACCTCATGGGGCTGAACTCAGCCGACCTGCTCAAGGGGCTG 60 (SEQ ID NO: 157)
E A D K S A Y L M G L N S A D L L K G L (SEQ ID NO: 155)
C H P Q V K V G N E Y V T K G Q S (SEQ ID NO: 96)
Mouse TGTCACCCTCAGGTGAAGGTGGGGAACGAGTATGTCACCAAGGGGCAGAGT 111 (SEQ ID NO: 156)
Human TGCCACCCTCAGGTGAAAGTGGGCAATGAGTACGTCACCAAGGGGCAGAAT 111 (SEQ ID NO: 157)
C H P Q V K V G N E Y V T K G Q N (SEQ ID NO: 155)
c.1208G>A

Fig. 10

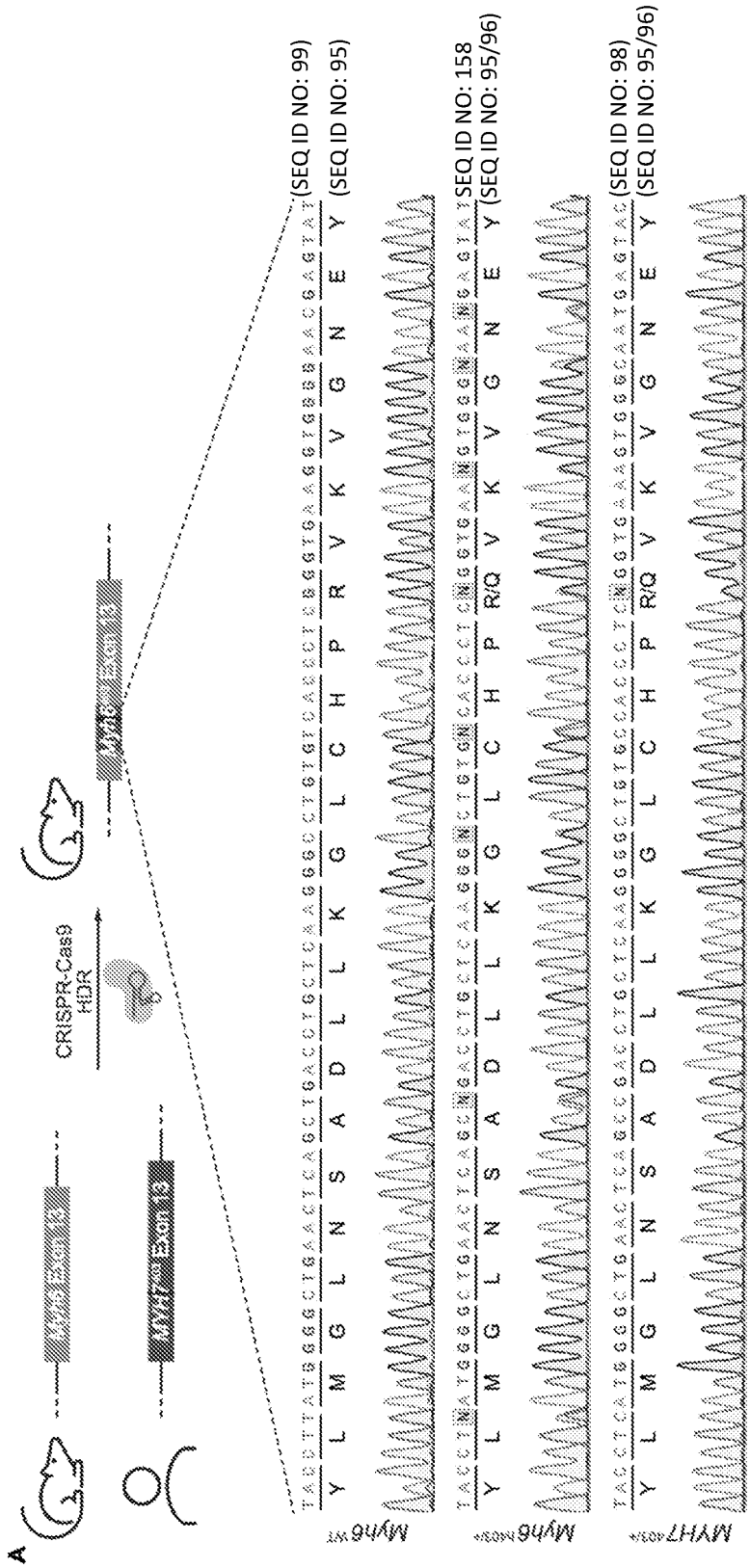

A
CRISPR-Cas9
HDR
Myh6 Exon 5
Myh7 Exon 5
Myh7 Exon 6
Myh6WT
Myh6NQSA
MYH7NQSA
(SEQ ID NO: 99)
(SEQ ID NO: 95)
SEQ ID NO: 158
(SEQ ID NO: 95/96)
(SEQ ID NO: 98)
(SEQ ID NO: 95/96)

Fig. 11A

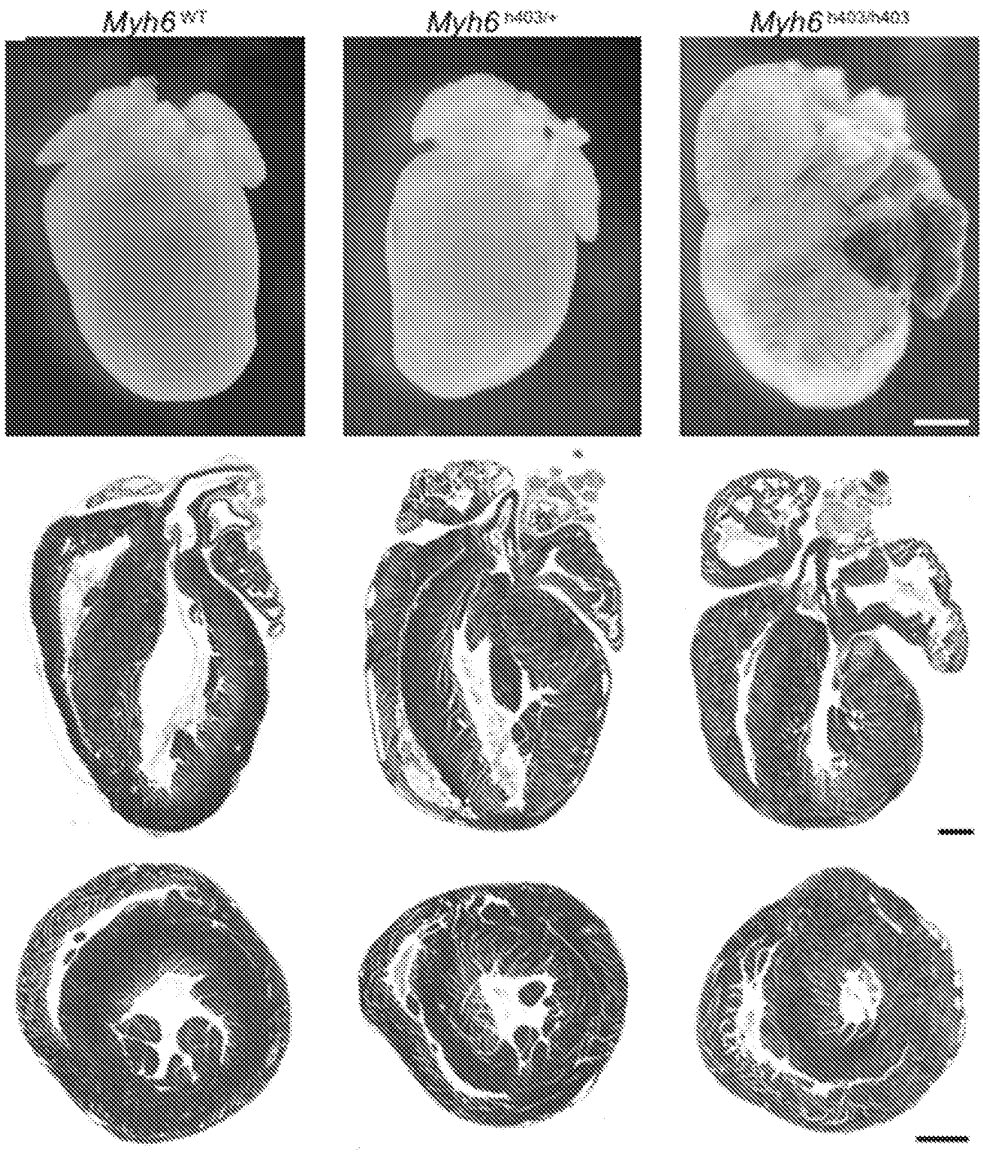


Fig. 11B

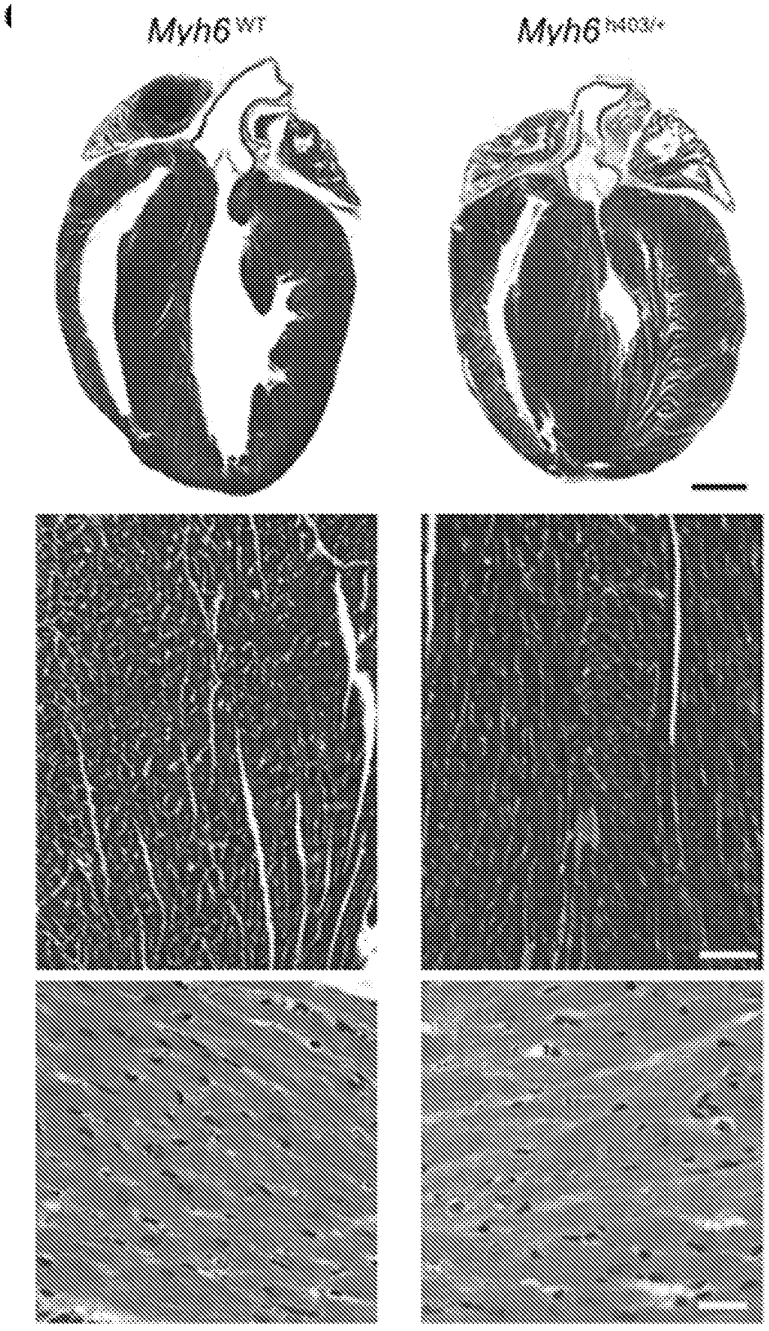


Fig. 11C

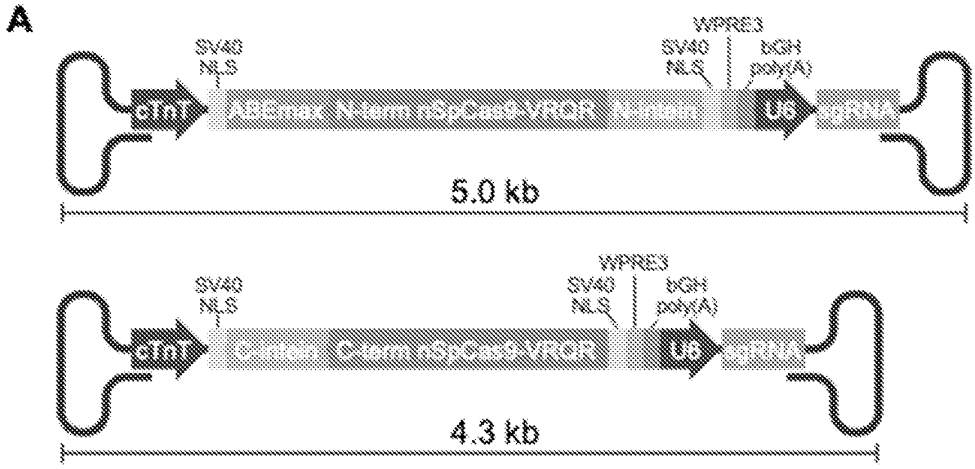


Fig. 12A

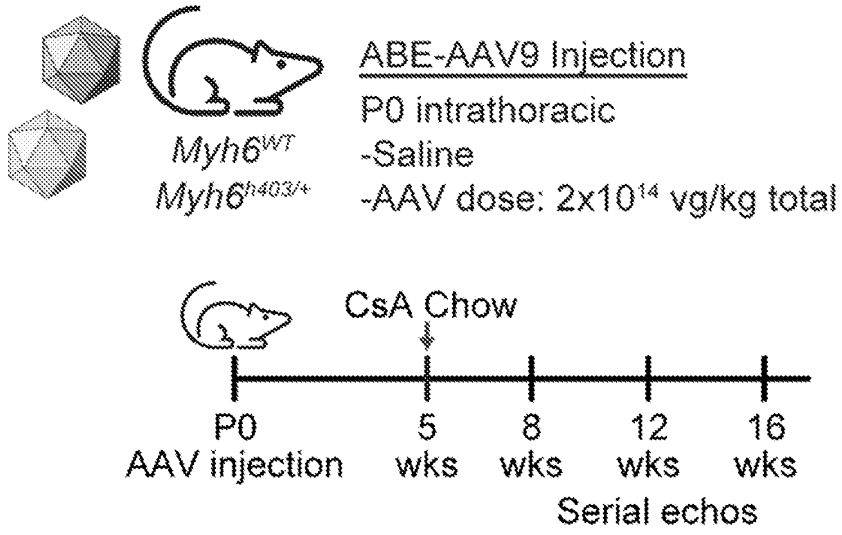


Fig. 12B

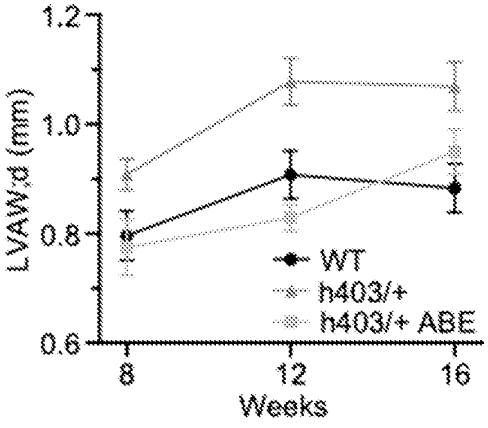


Fig. 12C

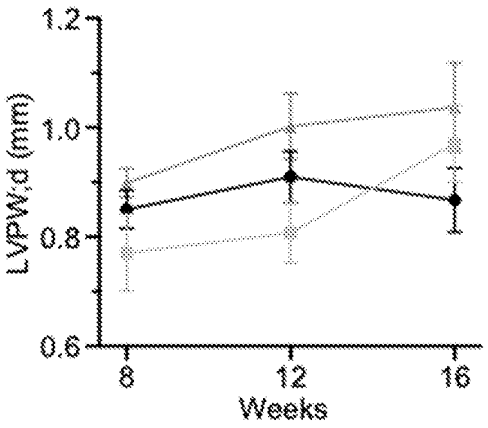


Fig. 12D

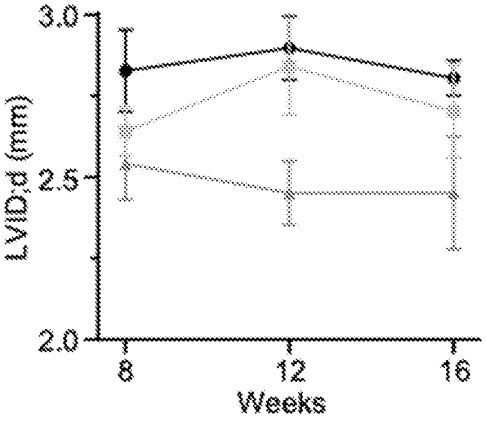


Fig. 12E

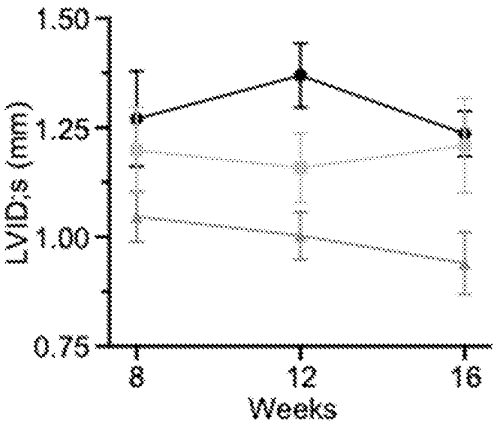


Fig. 12F

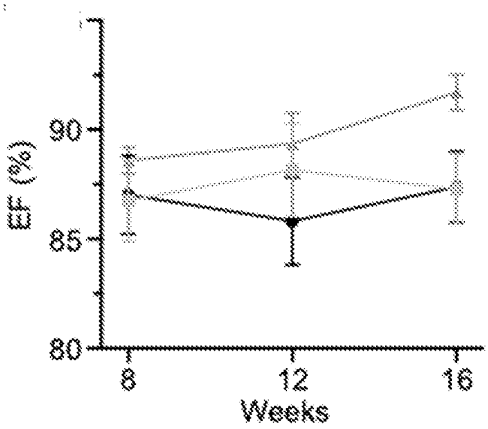


Fig. 12G

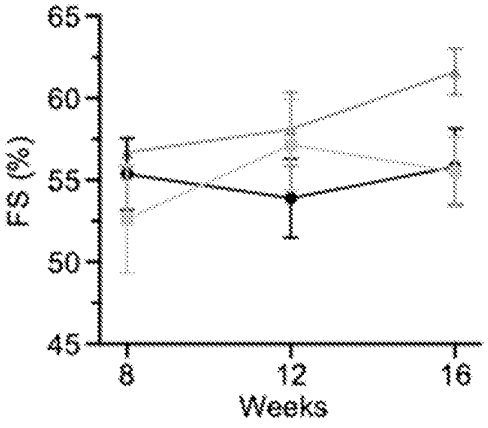


Fig. 12H

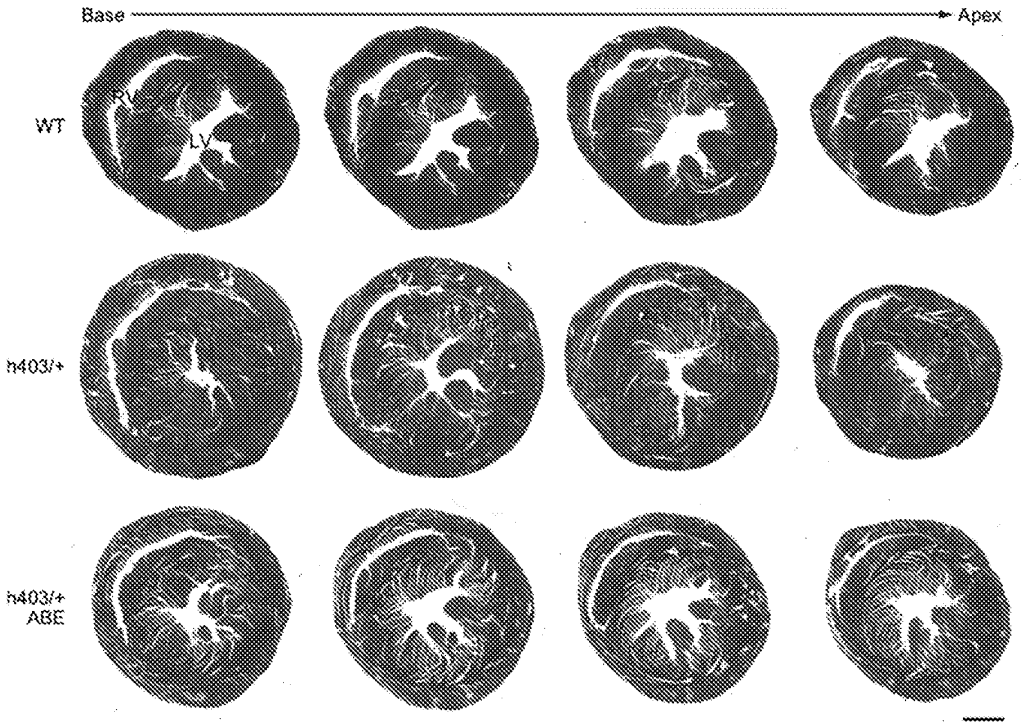


Fig. 12I

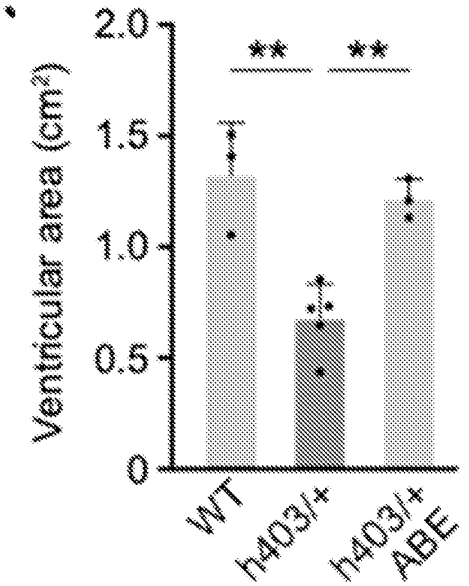


Fig. 12J

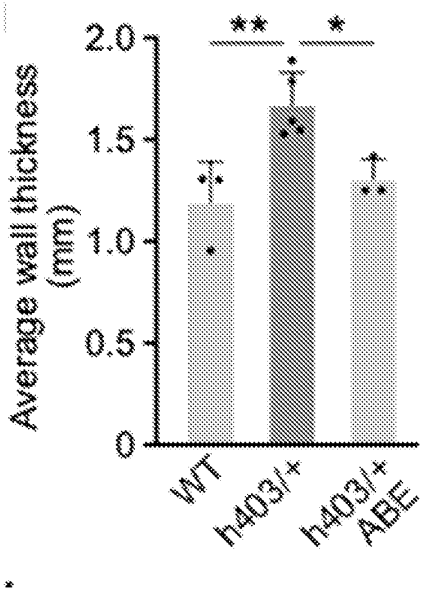


Fig. 12K

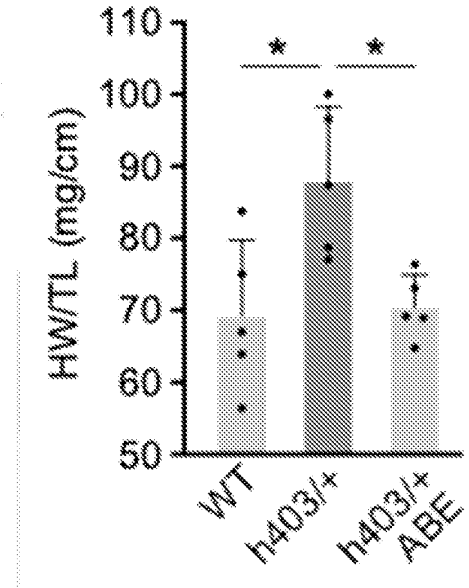


Fig. 12L

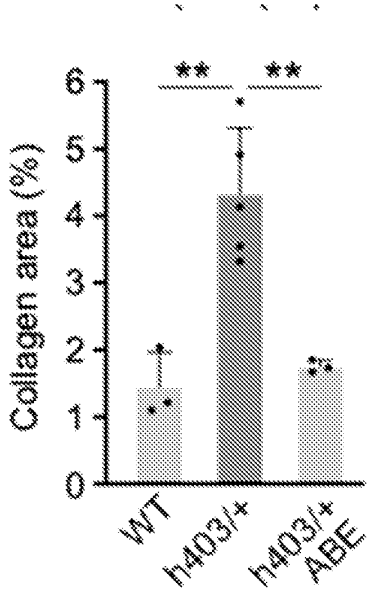
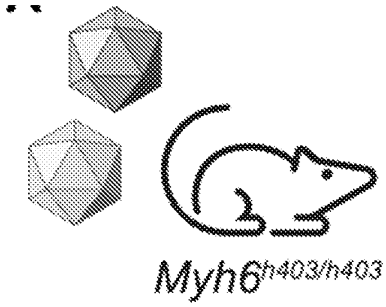


Fig. 12M



ABE-AAV9 Injection
P0 intrathoracic
-Saline
-AAV LOW: $8x10^{13}$ vg/kg total
-AAV HI: $3x10^{14}$ vg/kg total

Fig. 13A

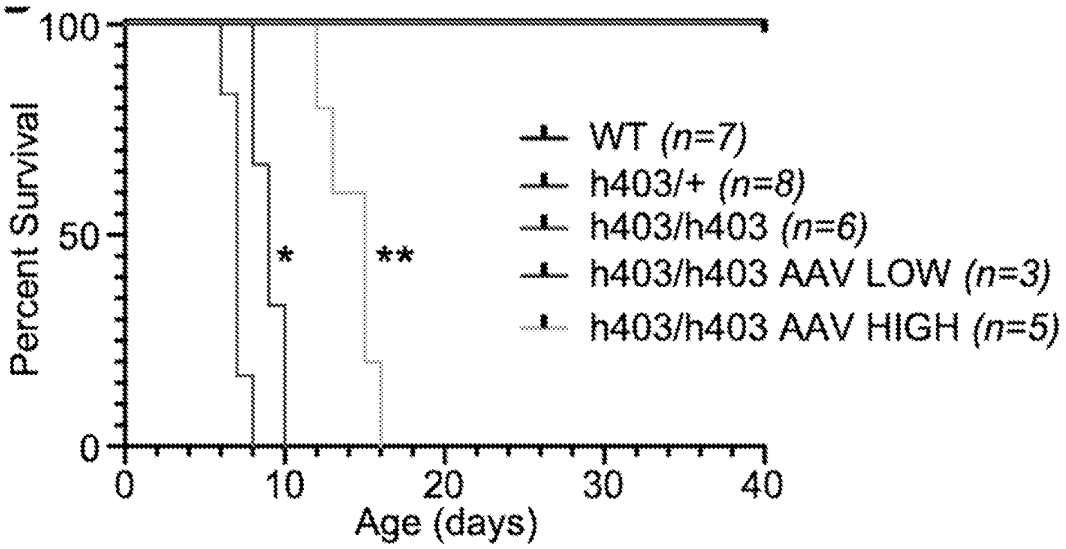


Fig. 13B

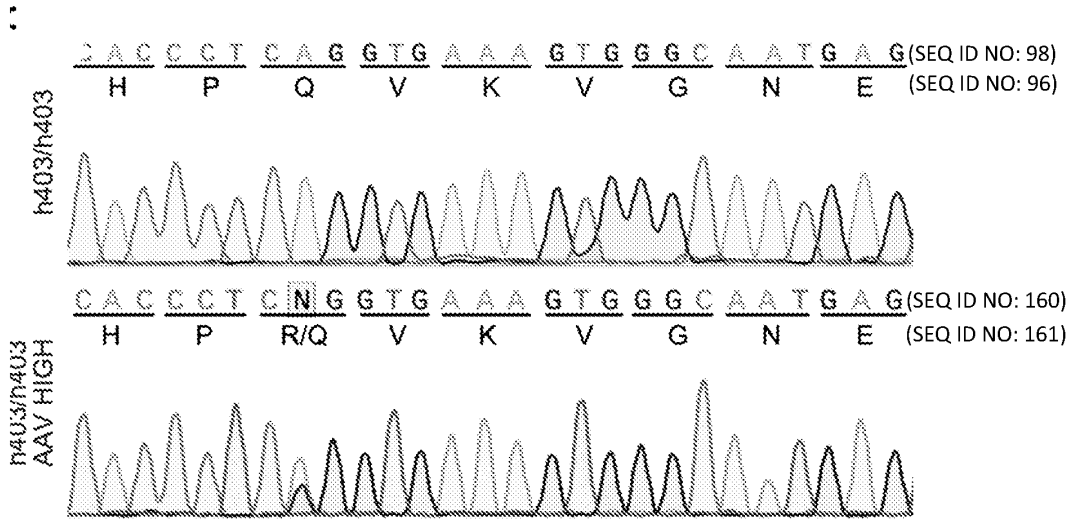


Fig. 13C

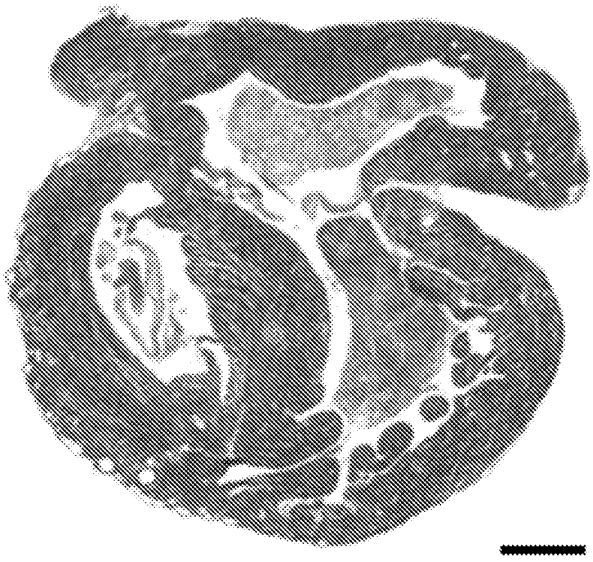


Fig. 13D

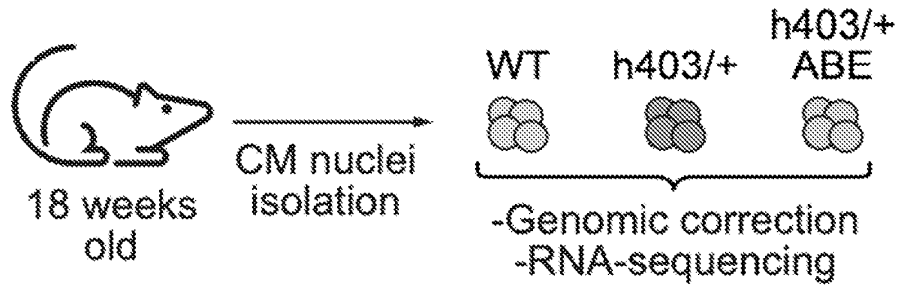


Fig. 14A

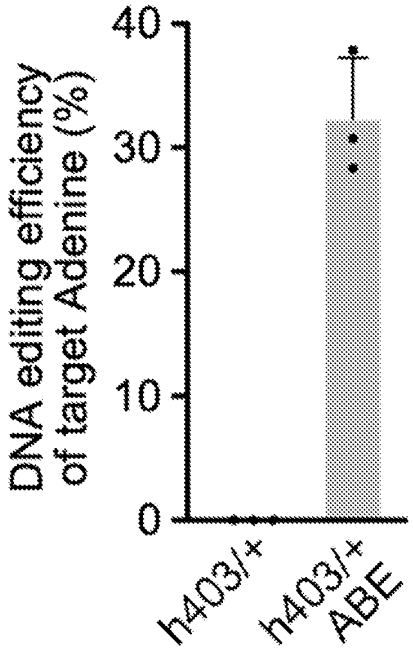


Fig. 14B

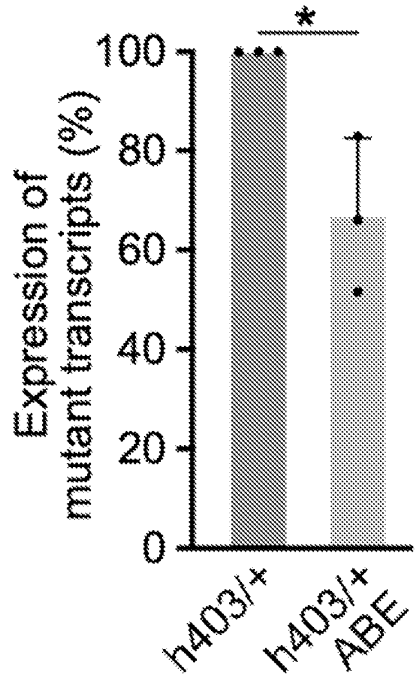


Fig. 14C

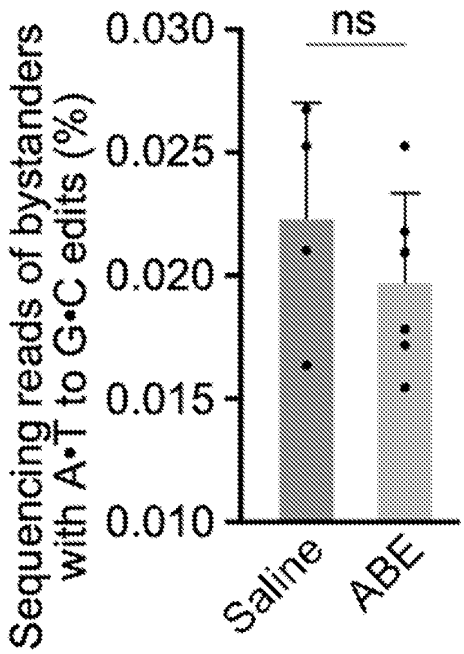


Fig. 14D

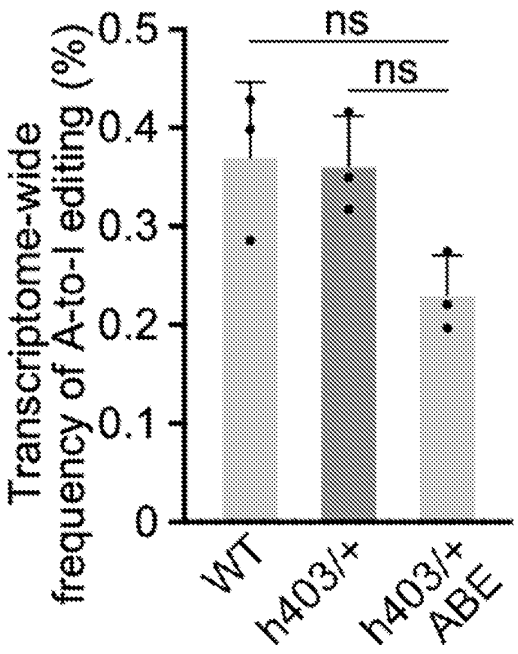


Fig. 14E

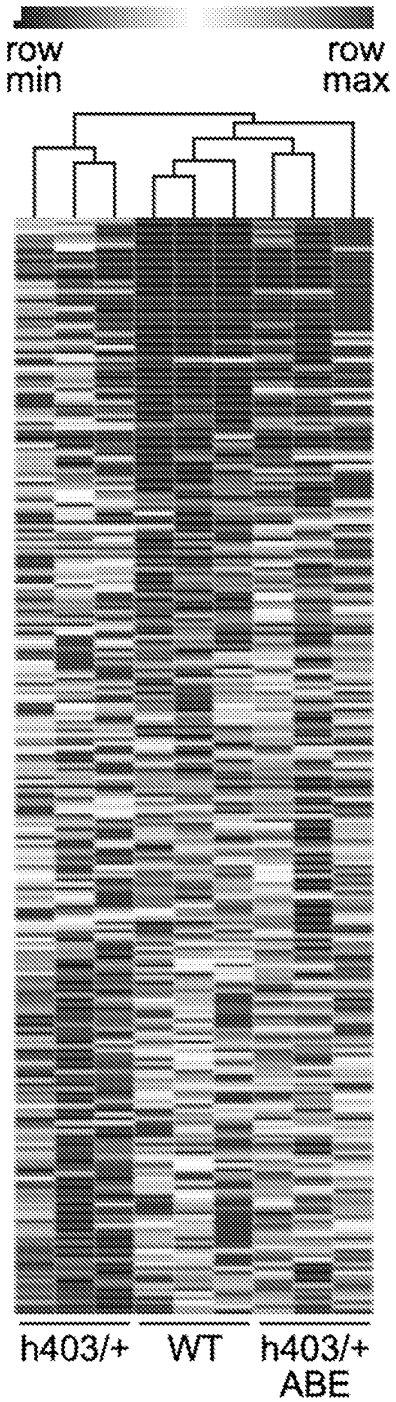


Fig. 14F

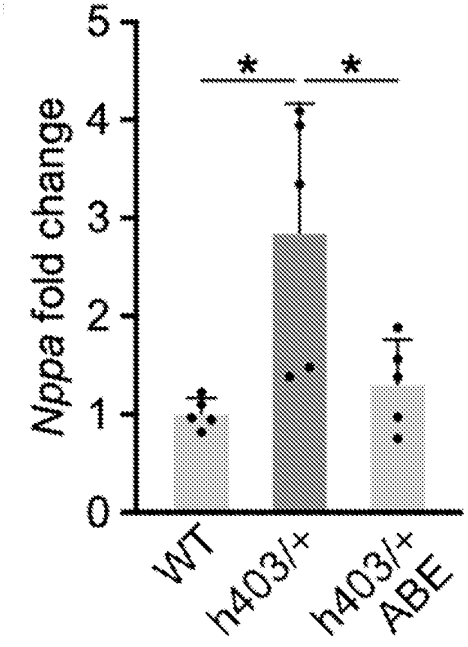


Fig. 14G

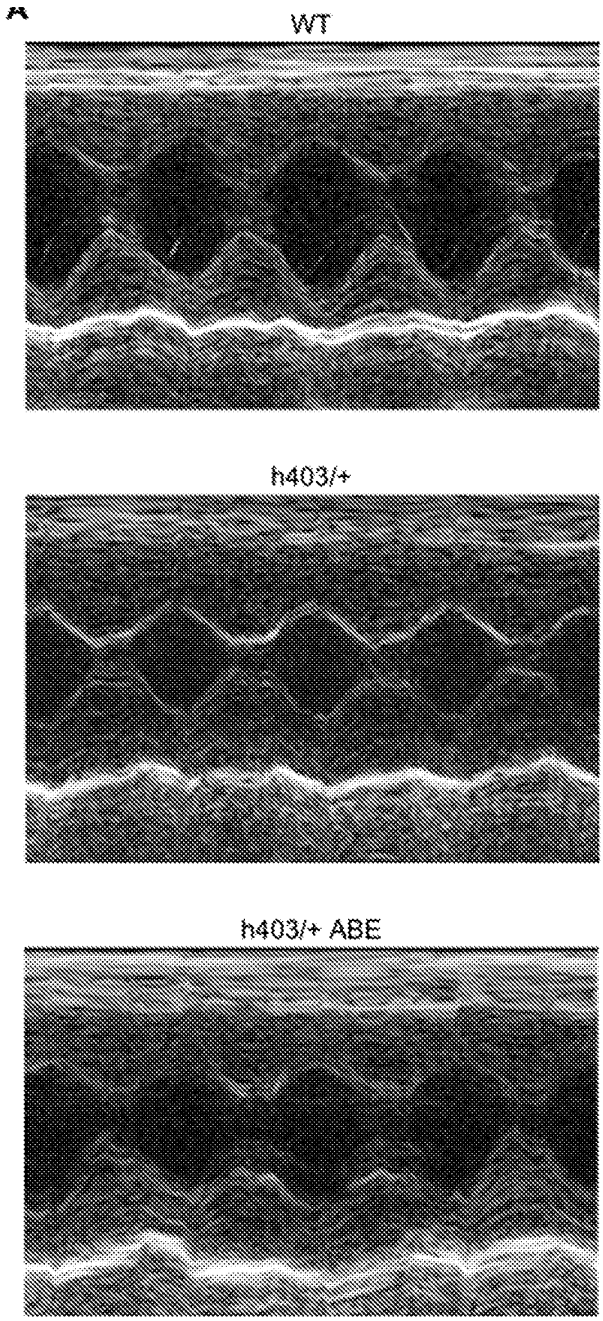


Fig. 15A

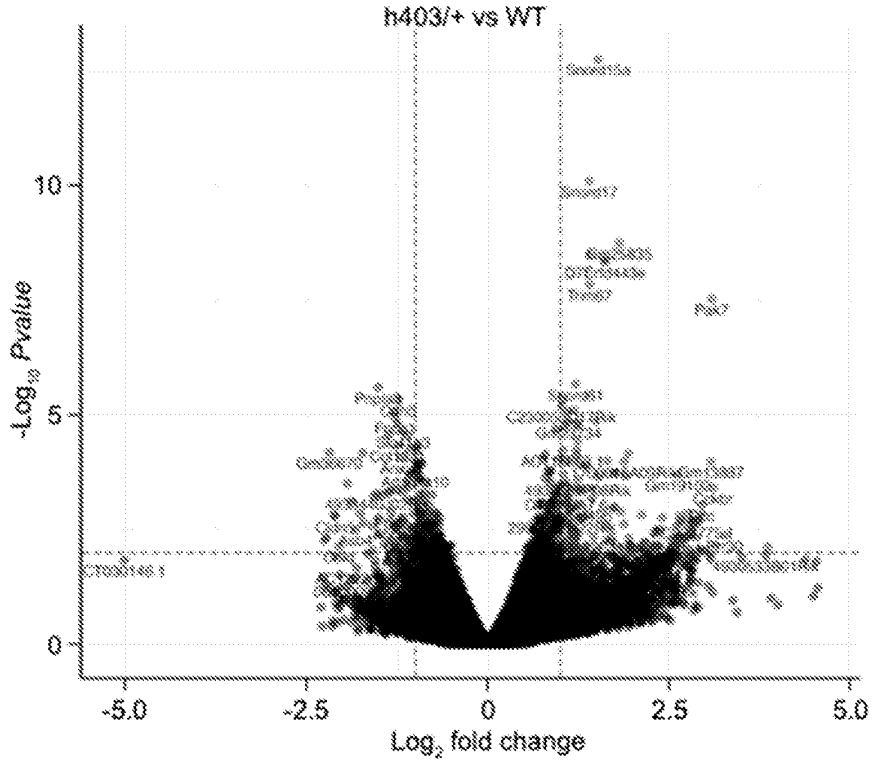


Fig. 15B

f

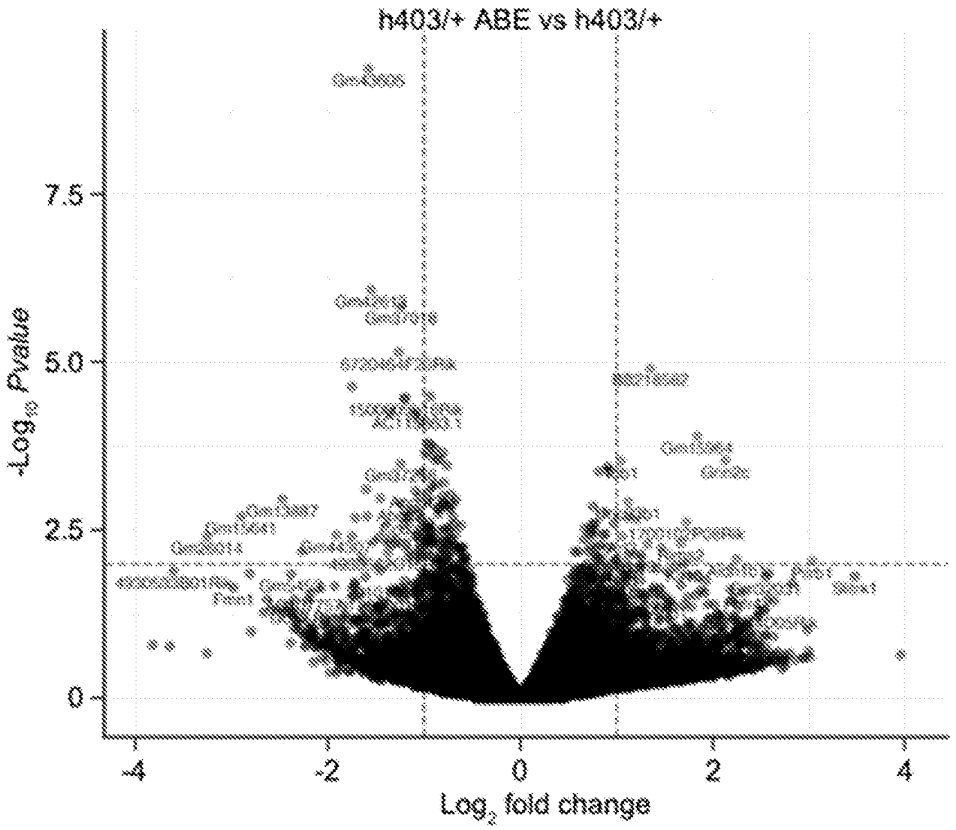

h403/+ ABE vs h403/+
7.5
5.0
2.5
0
-Log10 Pvalue
-4
-2
0
2
4
Log2 fold change

Fig. 15C

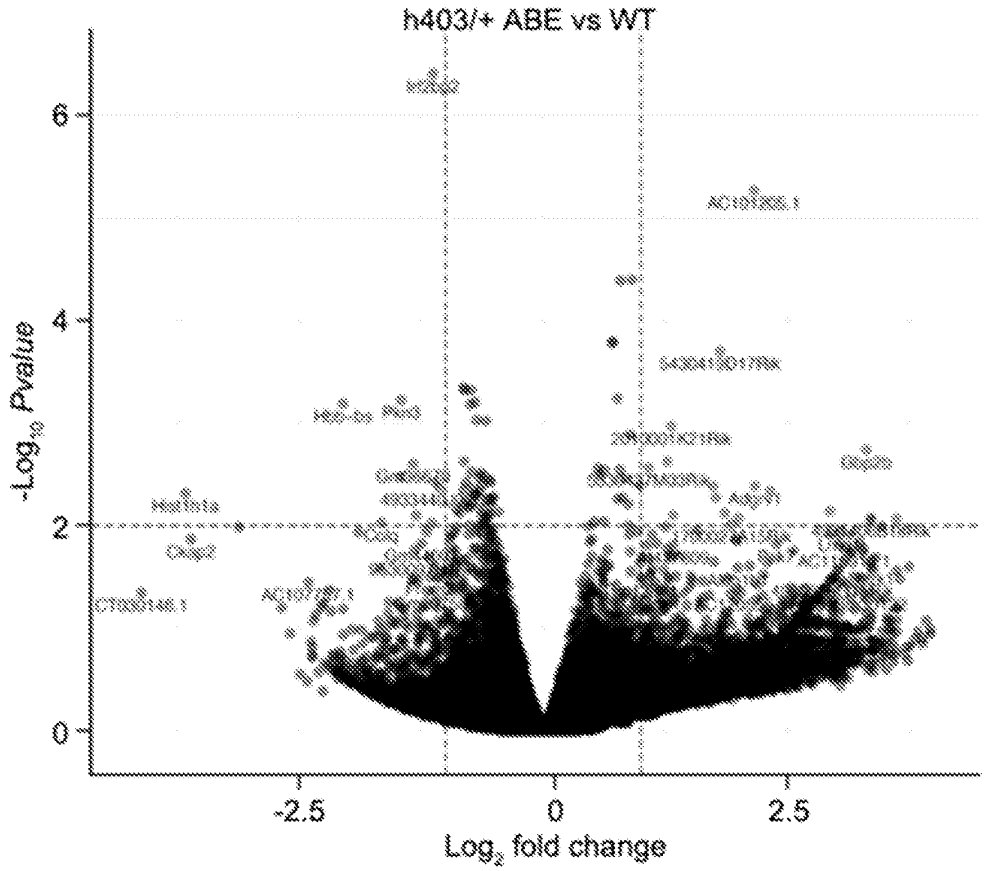


Fig. 15D