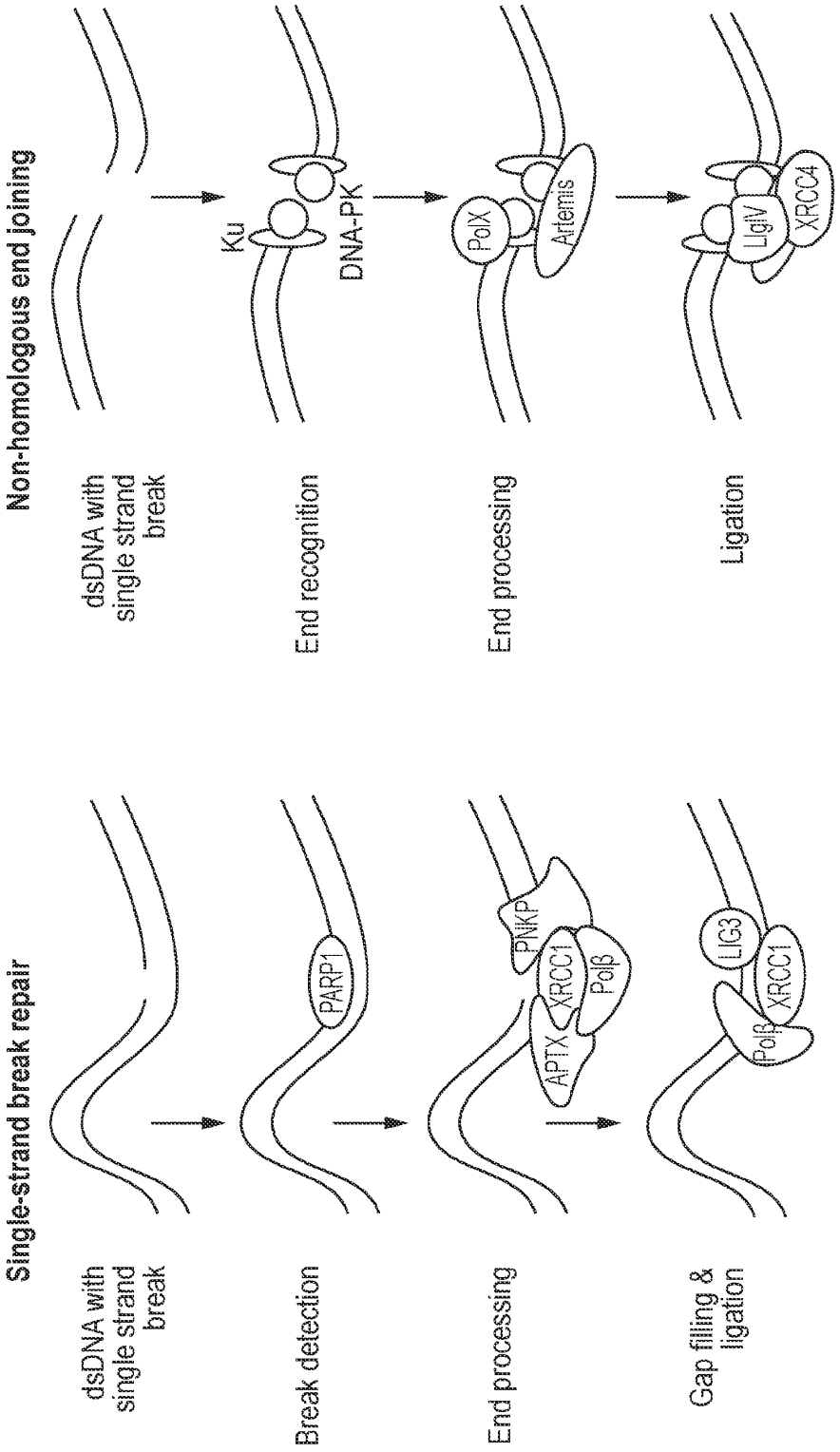

Non-homologous end joining
dsDNA with single strand break
Ku
DNA-PK
End recognition
PolX
Artemis
End processing
LigIV
XRCC4
Ligation
Single-strand break repair
dsDNA with single strand break
PARP1
Break detection
PNKP
XRCC1
Polβ
APTX
End processing
LIG3
XRCC1
Polβ
Gap filling & ligation

FIG. 1B

FIG. 1A

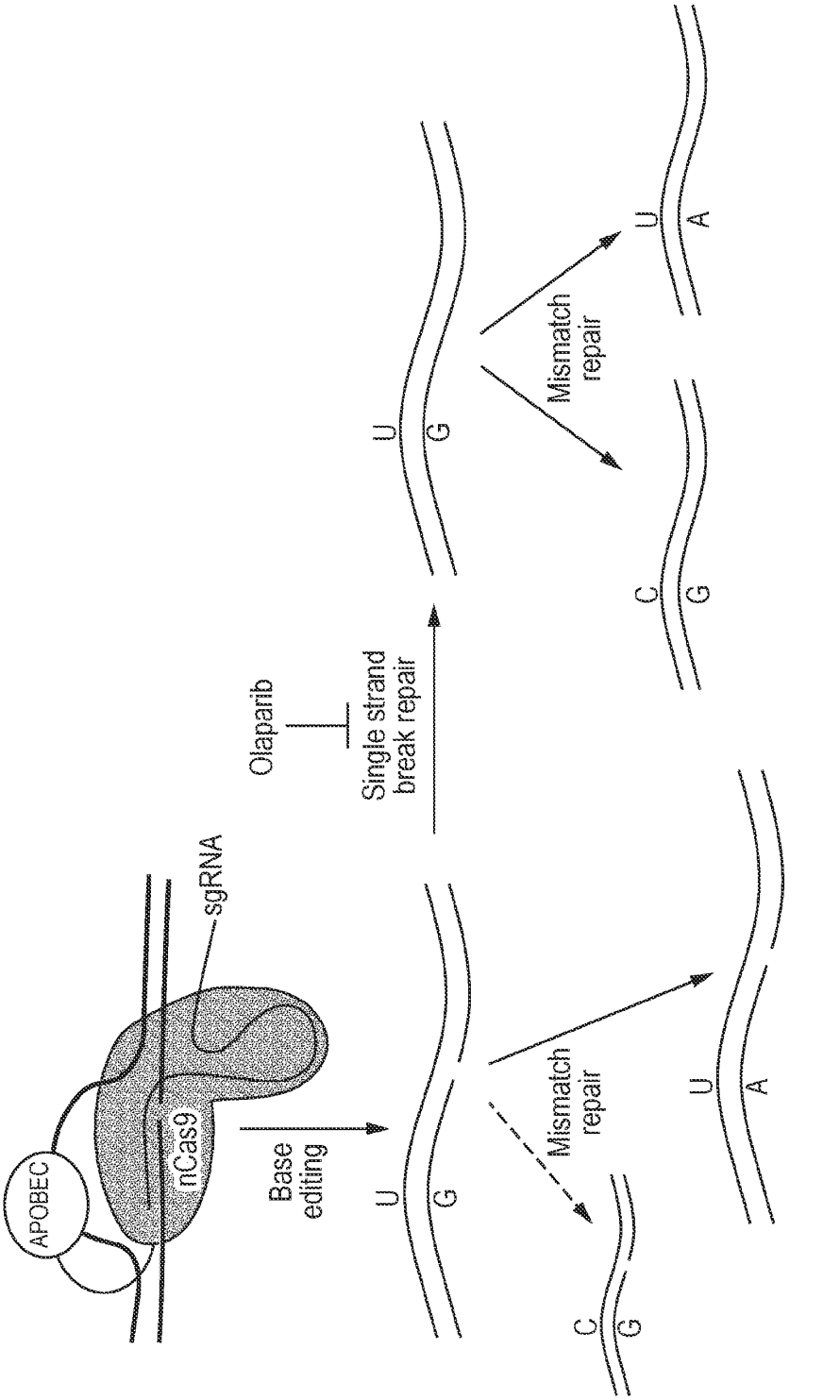

APOBEC
nCas9
sgRNA
Base editing
U
G
C
G
Mismatch repair
U
A
Olaparib
Single strand break repair
U
G
C
G
Mismatch repair
U
A

FIG. 2

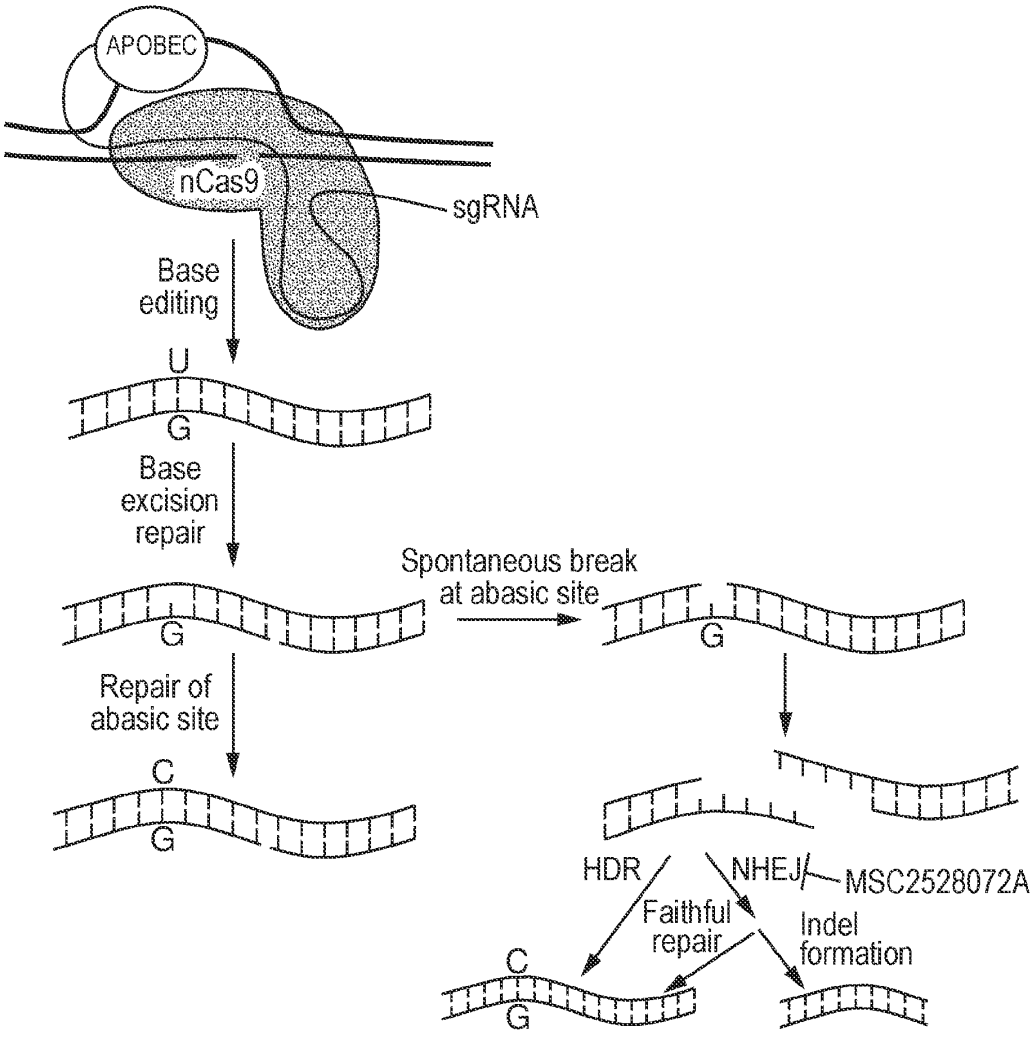

APOBEC
nCas9
sgRNA
Base editing
U
G
Base excision repair
G
Spontaneous break at abasic site
G
Repair of abasic site
C
G
HDR
NHEJ
MSC2528072A
Faithful repair
Indel formation
C
G

FIG. 3

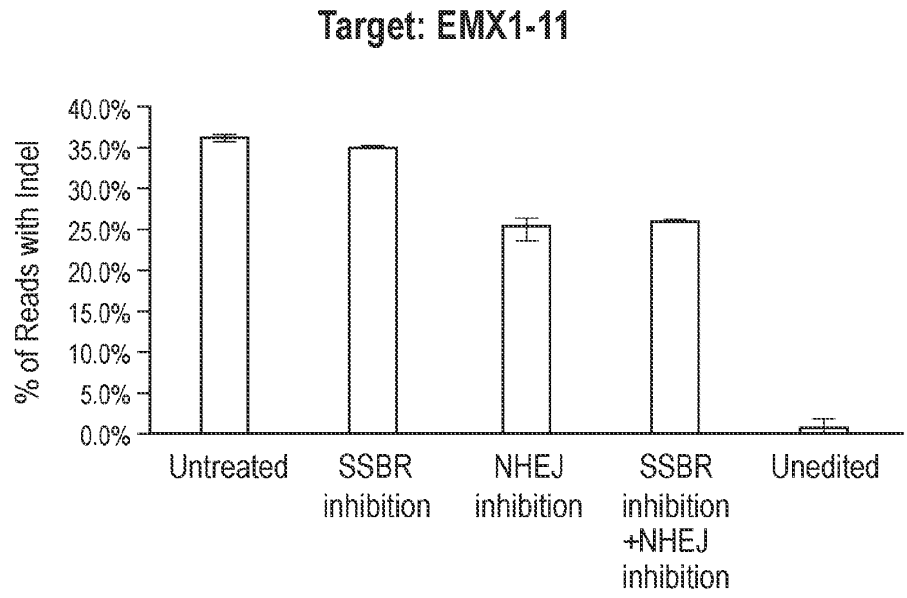

Target: EMX1-11
% of Reads with Indel
40.0%
35.0%
30.0%
25.0%
20.0%
15.0%
10.0%
5.0%
0.0%
Untreated
SSBR inhibition
NHEJ inhibition
SSBR inhibition +NHEJ inhibition
Unedited

FIG. 4A

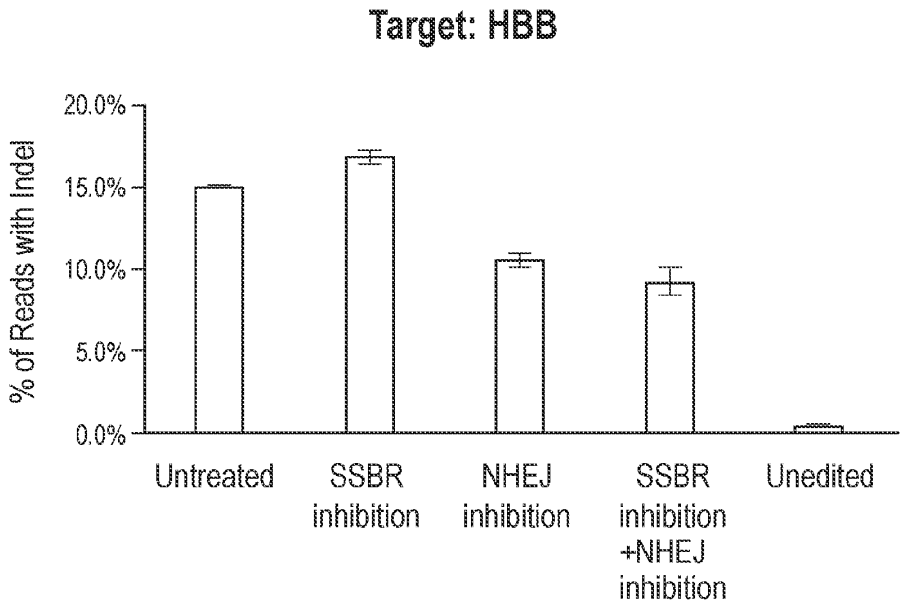

Target: HBB
% of Reads with Indel
20.0%
15.0%
10.0%
5.0%
0.0%
Untreated
SSBR inhibition
NHEJ inhibition
SSBR inhibition +NHEJ inhibition
Unedited

FIG. 4B

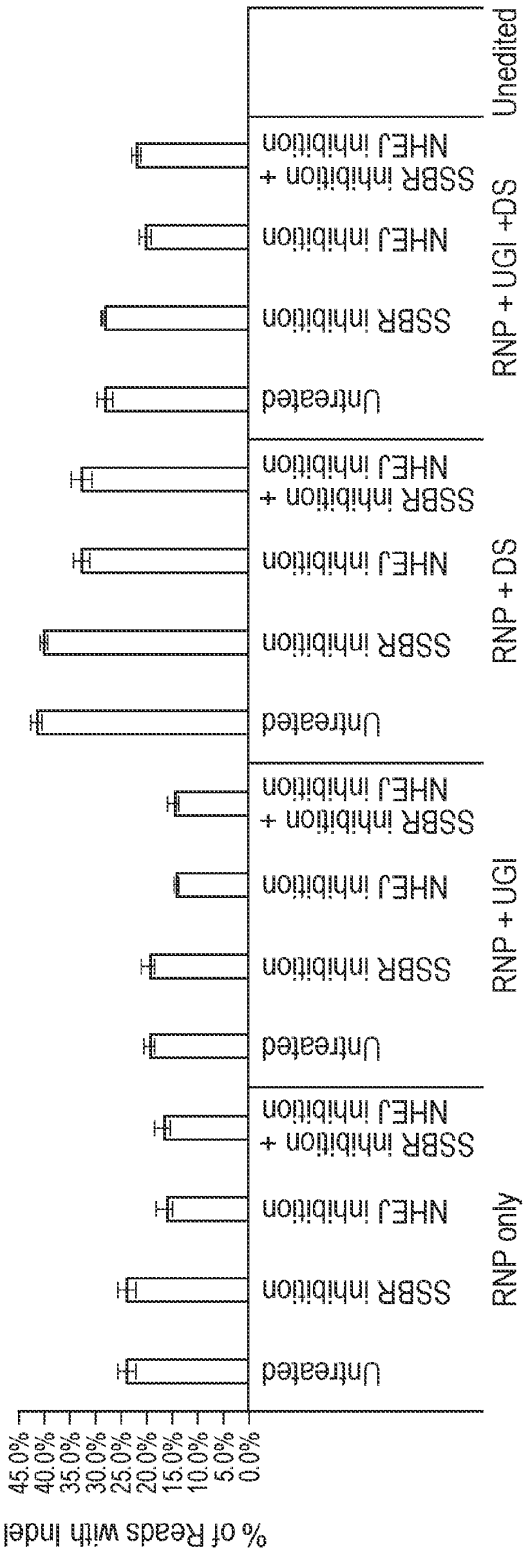

% of Reads with Indel
45.0%
40.0%
35.0%
30.0%
25.0%
20.0%
15.0%
10.0%
5.0%
0.0%
Untreated
SSBR inhibition
NHEJ inhibition
SSBR inhibition + NHEJ inhibition
Untreated
SSBR inhibition
NHEJ inhibition
SSBR inhibition + NHEJ inhibition
Untreated
SSBR inhibition
NHEJ inhibition
SSBR inhibition + NHEJ inhibition
Untreated
SSBR inhibition
NHEJ inhibition
SSBR inhibition + NHEJ inhibition
Unedited
RNP only
RNP + UGI
RNP + DS
RNP + UGI +DS

FIG. 5

SEQ ID NO: 1

APOBEC3B-SpCas9 nickase

GPGSEILRYLMDPDTFTFNFNNDPLVLRRRQTYLCYEVERLDNGTWVLMDQHMGFLCNEAKNLLCGFYGRHAELRFL
DLVPSLQLDPAQIYRVTWFISWSPCFSWGCAGEVRAFLQENTHVRLRIFAARIYDYDPLYKEALQMLRDAGAQVSIM
TYDEFEYCWDTFVYRQGCPFQPWDGLEEHSQALSGRLRAILQASSGGSSGGSSGSETPGTSESATPESSGGSSGGSP
AAKRVKLDGGGGSPKKKRKVTGMDKKYSIGLAIGTNSVGWAVITDEYKVPSKKFKVLGNTDRHSIKKNLIGALLFDS
GETAEATRLKRTARRRYTRRKNRICYLQEIFSNEMAKVDDSFFHRLEESFLVEEDKKHERHPIFGNIVDEVAYHEKY
PTIYHLRKKLVDSTDKADLRLIYLALAHMIKFRGHFLIEGDLNPDNSDVDKLFIQLVQTYNQLFEENPINASGVDAK
AILSARLSKSRRLENLIAQLPGEKKNGLFGNLIALSLGLTPNFKSNFDLAEDAKLQLSKDTYDDDLDNLLAQIGDQY
ADLFLAAKNLSDAILLSDILRVNTEITKAPLSASMIKRYDEHHQDLTLLKALVRQQLPEKYKEIFFDQSKNGYAGYI
DGGASQEEFYKFIKPILEKMDGTEELLVKLNREDLLRKQRTFDNGSIPHQIHLGELHAILRRQEDFYPFLKDNREKI
EKILTFRIPYYVGPLARGNSRFAWMTRKSEETITPWNFEEVVDKGASAQSFIERMTNFDKNLPNEKVLPKHSLLYEY
FTVYNELTKVKYVTEGMRKPAFLSGEQKKAIVDLLFKTNRKVTVKQLKEDYFKKIECFDSVEISGVEDRFNASLGTY
HDLLKIIKDKDFLDNEENEDILEDIVLTLTLFEDREMIEERLKTYAHLFDDKVMKQLKRRRYTGWGRLSRKLINGIR
DKQSGKTILDFLKSDGFANRNFMQLIHDDSLTFKEDIQKAQVSGQGDSLHEHIANLAGSPAIKKGILQTVKVVDELV
KVMGRHKPENIVIEMARENQTTQKGQKNSRERMKRIEEGIKELGSQILKEHPVENTQLQNEKLYLYYLQNGRDMYVD
QELDINRLSDYDVDHIVPQSFLKDDSIDNKVLTRSDKNRGKSDNVPSEEVVKKMKNYWRQLLNAKLITQRKFDNLTK
AERGGLSELDKAGFIKRQLVETRQITKHVAQILDSRMNTKYDENDKLIREVKVITLKSKLVSDFRKDFQFYKVREIN
NYHHAHDAYLNAVVGTALIKKYPKLESEFVYGDYKVYDVRKMIAKSEQEIGKATAKYFFYSNIMNFFKTEITLANGE
IRKRPLIETNGETGEIVWDKGRDFATVRKVLSMPQVNIVKKTEVQTGGFSKESILPKRNSDKLIARKKDWDPKKYGG
FDSPTVAYSVLVVAKVEKGKSKKLKSVKELLGITIMERSSFEKNPIDFLEAKGYKEVKKDLIIKLPKYSLFELENGR
KRMLASAGELQKGNELALPSKYVNFLYLASHYEKLKGSPEDNEQKQLFVEQHKHYLDEIIEQISEFSKRVILADANL
DKVLSAYNKHRDKPIREQAENIIHLFTLTNLGAPAAFKYFDTTIDRKRYTSTKEVLDATLIHQSITGLYETRIDLSQ
LGGDEFPKKKRKVGGGGSPKKKRKV

SEQ ID NO: 2

UGI with NLS (Italic: linker; underline: NLS)

MTNLSDIIEKETGKQLVIQESILMLPEEVEEVIGNKPESDILVHTAYDESTDENVMLLTSDAPEYKPWALVIQDSNG
ENKIKML*SGGSGGSGGSGGSPAAKRVKLDGGGGSPKKKRKV*

FIG. 6

AAVS1 guide sequence	GGGGCCACTAGGGACAGGAT SEQ ID NO: 21
BCL4 guide sequence	TGTTGTGATTCCGAGCTCCG SEQ ID NO: 22
AAVS1 donor sequence	GTTCTTGGGTACTTTTTATCTTGTCCCCTCCACCCCACAGTGGGGCCACTAGGGACAGACCTAGGCCTGATTGGTGACAGAAAAGCCCATCCTTAGGCCTCCTTCCTTCCTAG SEQ ID NO: 23
BCL4 donor sequence	CTGCCCGCATCATTATGATGATAACTATTACTATTGTTGTGATTCCGAGTTGGTACCGGCCCCGAGGCGAGAGGGGGGTGTCGGGCTGCAGAGCCGCCAGCAAGCCAGC SEQ ID NO: 24
AAVS1 NGS F	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGNNNNNNNAGTTCTGGCAAGGAGAGATG SEQ ID NO: 25
AAVS1 NGS R	GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGNNNNNNCTATGTCCACTTCAGGACAGCATG SEQ ID NO: 26
BCL4 NGS F	TCGTCGGCAGCGTCAGATGTGTATAAGAGACAGNNNNNNGTCCAGCCTCACTTCTTCC SEQ ID NO: 27
BCL4 NGS R	GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAGNNNNNNGGTCCTGAGATTCATTCTGTGTCC SEQ ID NO: 28

FIG. 7

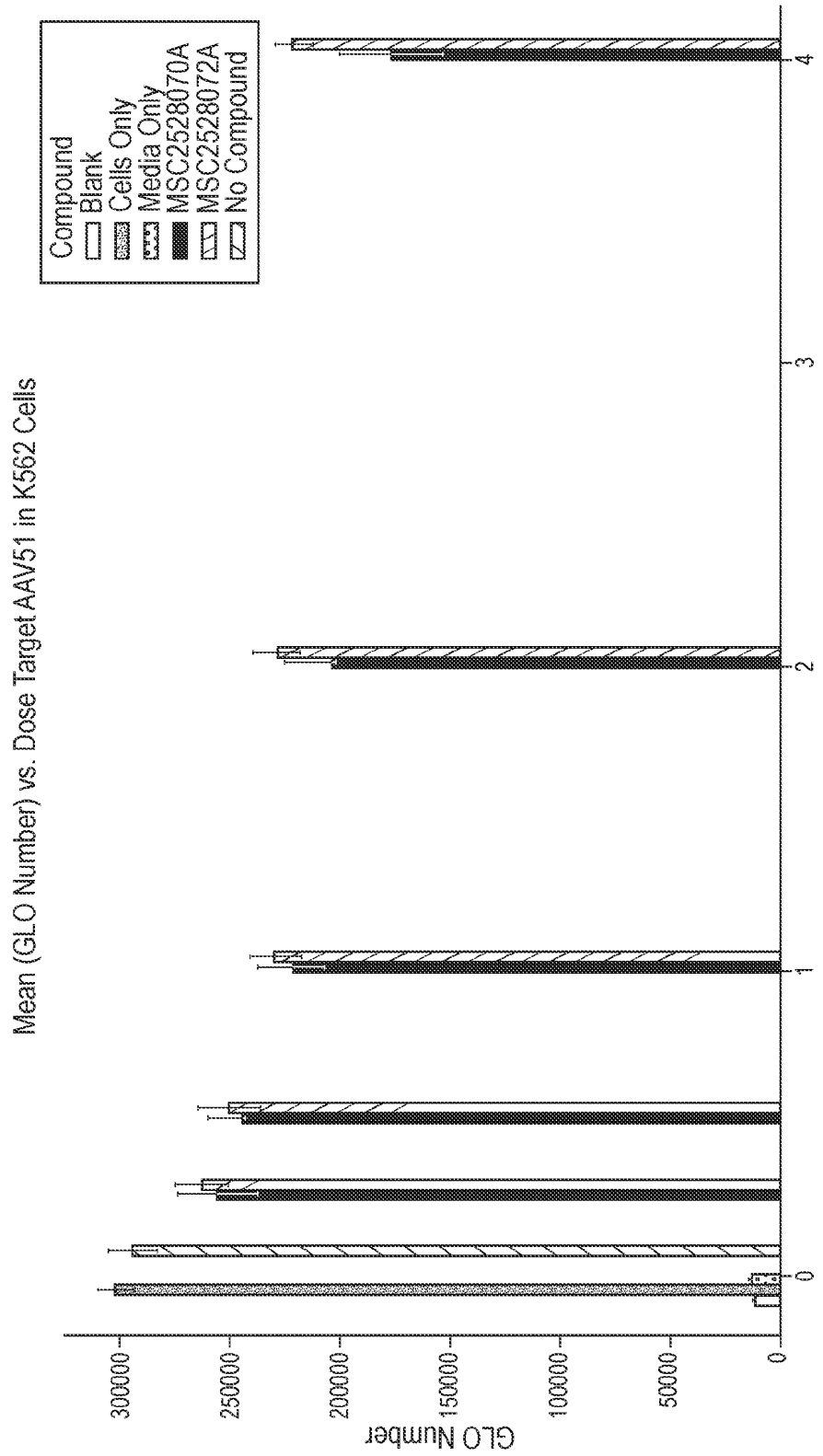

Mean (GLO Number) vs. Dose Target AAV51 in K562 Cells
Compound
Blank
Cells Only
Media Only
MSC2528070A
MSC2528072A
No Compound
GLO Number
300000
250000
200000
150000
100000
50000
0
0
1
2
3
4
Dose

FIG. 8

Compound	Replicate	Dose (μM)	GLO Number
MSC2528070A	2	4	337583
MSC2528072A	2	4	343653
MSC2528070A	1	4	325850
MSC2528072A	1	4	354580
MSC2528070A	2	2	334296
MSC2528072A	2	2	344707
MSC2528070A	1	2	326186
MSC2528072A	1	2	339531
MSC2528070A	2	1	338612
MSC2528072A	2	1	340510
MSC2528070A	1	1	323068
MSC2528072A	1	1	334431
MSC2528070A	2	0.5	323623
MSC2528072A	2	0.5	337702
MSC2528070A	1	0.5	326970
MSC2528072A	1	0.5	331349
MSC2528070A	2	0.25	338420
MSC2528072A	2	0.25	334415
MSC2528070A	1	0.25	320155
MSC2528072A	1	0.25	330852
Blank	2	0	11164
Media Only	2	0	16994
No Compound	2	0	331744
Cells Only	2	0	280599
Blank	1	0	17021
Media Only	1	0	15566
No Compound	1	0	329112
Cells Only	1	0	288637

FIG. 9

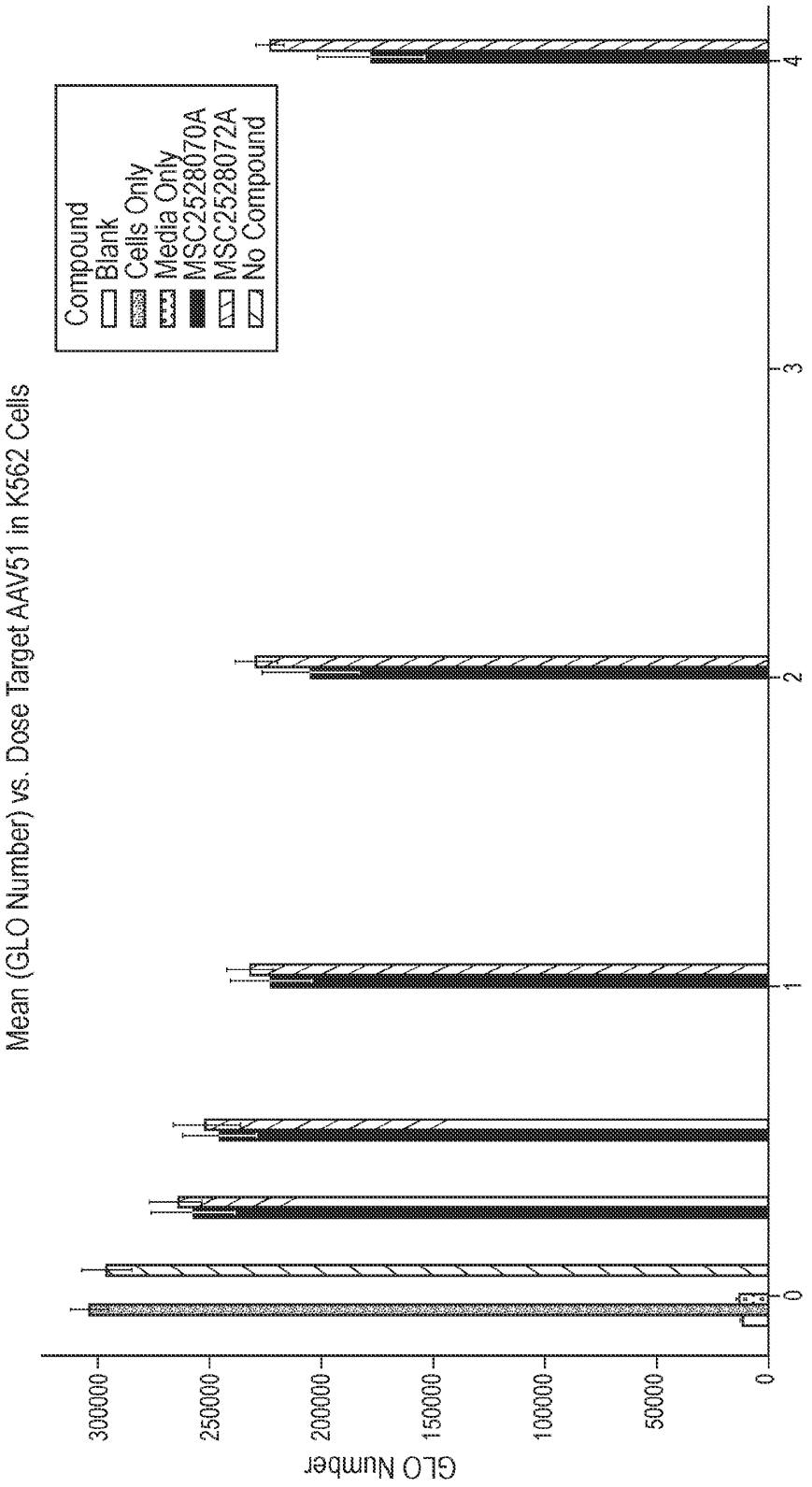

Mean (GLO Number) vs. Dose Target AAV51 in K562 Cells
GLO Number
300000
250000
200000
150000
100000
50000
0
0
1
2
3
4
Dose
Compound
Blank
Cells Only
Media Only
MSC2528070A
MSC2528072A
No Compound

FIG. 10

Compound	Replicate	Dose	GLO Number
MSC2528070A	2	4	199756
MSC2528072A	2	4	227513
MSC2528070A	1	4	152303
MSC2528072A	1	4	215066
MSC2528070A	2	2	224707
MSC2528072A	2	2	236001
MSC2528070A	1	2	180489
MSC2528072A	1	2	218671
MSC2528070A	2	1	238591
MSC2528072A	2	1	240091
MSC2528070A	1	1	202134
MSC2528072A	1	1	219137
MSC2528070A	2	0.5	259617
MSC2528072A	2	0.5	264005
MSC2528070A	1	0.5	226651
MSC2528072A	1	0.5	235224
MSC2528070A	2	0.25	273554
MSC2528072A	2	0.25	273635
MSC2528070A	1	0.25	236291
MSC2528072A	1	0.25	250159
Blank	2	0	8650
Media Only	2	0	13516
No Compound	2	0	304068
Cells Only	2	0	292864
Blank	1	0	11938
Media Only	1	0	11034
No Compound	1	0	282320
Cells Only	1	0	308742

FIG. 11

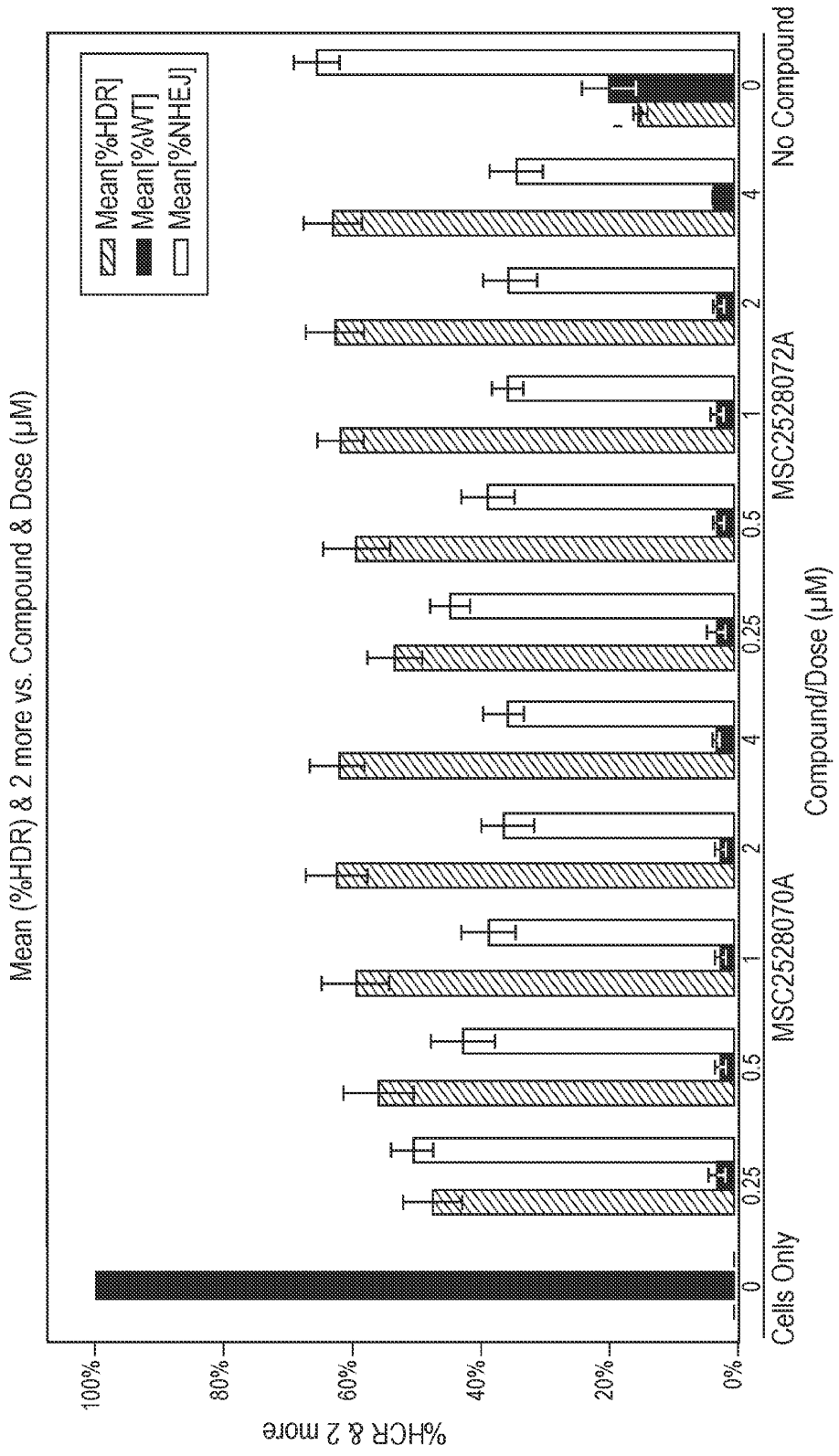

Mean (%HDR) & 2 more vs. Compound & Dose (µM)
Mean[%HDR]
Mean[%WT]
Mean[%NHEJ]
%HCR & 2 more
100%
80%
60%
40%
20%
0%
Cells Only
MSC2528070A
MSC2528072A
No Compound
Compound/Dose (µM)
0
0.25
0.5
1
2
4
0.25
0.5
1
2
4
0

FIG. 12

Target	Compound	Replicate	Dose (μM)	Wash	GLO #	%HDR	%WT	%NHEJ
AAVS1	MSC2528070A	1	0.25	0	236291	50.31%	1.70%	47.99%
AAVS1	MSC2528070A	1	0.25	1	231903	41.62%	6.20%	52.18%
AAVS1	MSC2528070A	1	0.5	0	226651	59.47%	1.60%	38.93%
AAVS1	MSC2528070A	1	0.5	1	223873	47.01%	4.70%	48.29%
AAVS1	MSC2528070A	1	1	0	202134	62.99%	1.70%	35.31%
AAVS1	MSC2528070A	1	1	1	219443	49.02%	3.70%	47.28%
AAVS1	MSC2528070A	1	2	0	180489	65.45%	1.70%	32.85%
AAVS1	MSC2528070A	1	2	1	185814	56.94%	1.60%	41.46%
AAVS1	MSC2528070A	1	4	0	152303	65.03%	2.60%	32.37%
AAVS1	MSC2528070A	1	4	1	205750	54.21%	1.90%	43.89%
AAVS1	MSC2528070A	2	0.25	0	273554	43.71%	3.60%	52.69%
AAVS1	MSC2528070A	2	0.25	1	231727	38.04%	8.50%	53.46%
AAVS1	MSC2528070A	2	0.5	0	259617	51.46%	2.70%	45.84%
AAVS1	MSC2528070A	2	0.5	1	244978	44.28%	6.40%	49.32%
AAVS1	MSC2528070A	2	1	0	238591	55.64%	2.80%	41.56%
AAVS1	MSC2528070A	2	1	1	246267	46.67%	4.80%	48.53%
AAVS1	MSC2528070A	2	2	0	224707	58.74%	2.80%	38.46%
AAVS1	MSC2528070A	2	2	1	235200	43.04%	5.60%	51.36%
AAVS1	MSC2528070A	2	4	0	199756	58.60%	3.30%	38.10%
AAVS1	MSC2528070A	2	4	1	238017	50.76%	3.10%	46.14%
AAVS1	MSC2528072A	1	0.25	0	250159	55.88%	1.80%	42.32%

FIG. 13

AAVS1	MSC2528072A	1	0.25	1	216718	44.79%	5.10%	50.11%
AAVS1	MSC2528072A	1	0.5	0	235224	62.81%	2.00%	35.19%
AAVS1	MSC2528072A	1	0.5	1	228164	49.02%	4.10%	46.88%
AAVS1	MSC2528072A	1	1	0	219137	64.19%	2.10%	33.71%
AAVS1	MSC2528072A	1	1	1	231591	50.12%	3.50%	46.38%
AAVS1	MSC2528072A	1	2	0	218671	65.64%	2.20%	32.16%
AAVS1	MSC2528072A	1	2	1	200751	58.59%	2.00%	39.41%
AAVS1	MSC2528072A	1	4	0	215066	66.06%	3.20%	30.74%
AAVS1	MSC2528072A	1	4	1	200083	62.21%	2.00%	35.79%
AAVS1	MSC2528072A	2	0.25	0	273635	49.83%	3.70%	46.47%
AAVS1	MSC2528072A	2	0.25	1	254920	41.06%	8.50%	50.44%
AAVS1	MSC2528072A	2	0.5	0	264005	55.35%	3.30%	41.35%
AAVS1	MSC2528072A	2	0.5	1	244012	45.04%	6.00%	48.96%
AAVS1	MSC2528072A	2	1	0	240091	59.20%	3.60%	37.20%
AAVS1	MSC2528072A	2	1	1	241568	46.68%	5.10%	48.22%
AAVS1	MSC2528072A	2	2	0	236001	58.84%	3.30%	37.86%
AAVS1	MSC2528072A	2	2	1	233297	50.58%	4.20%	45.22%
AAVS1	MSC2528072A	2	4	0	227513	59.69%	3.50%	36.81%
AAVS1	MSC2528072A	2	4	1	232133	55.50%	3.20%	41.30%
AAVS1	Cells Only	2	0	1	324596	0.00%	100.00%	0.00%
AAVS1	Cells Only	1	0	1	362811	0.00%	100.00%	0.00%
AAVS1	Cells Only	2	0	0	292864	0.00%	100.00%	0.00%
AAVS1	Cells Only	1	0	0	308742	0.00%	100.00%	0.00%
AAVS1	No Compound	2	0	1	285973	16.30%	23.10%	60.60%
AAVS1	No Compound	1	0	1	260796	16.09%	15.80%	68.11%
AAVS1	No Compound	2	0	0	304068	14.89%	22.50%	62.61%
AAVS1	No Compound	1	0	0	282320	15.62%	16.50%	67.88%

FIG. 13 (Cont.)

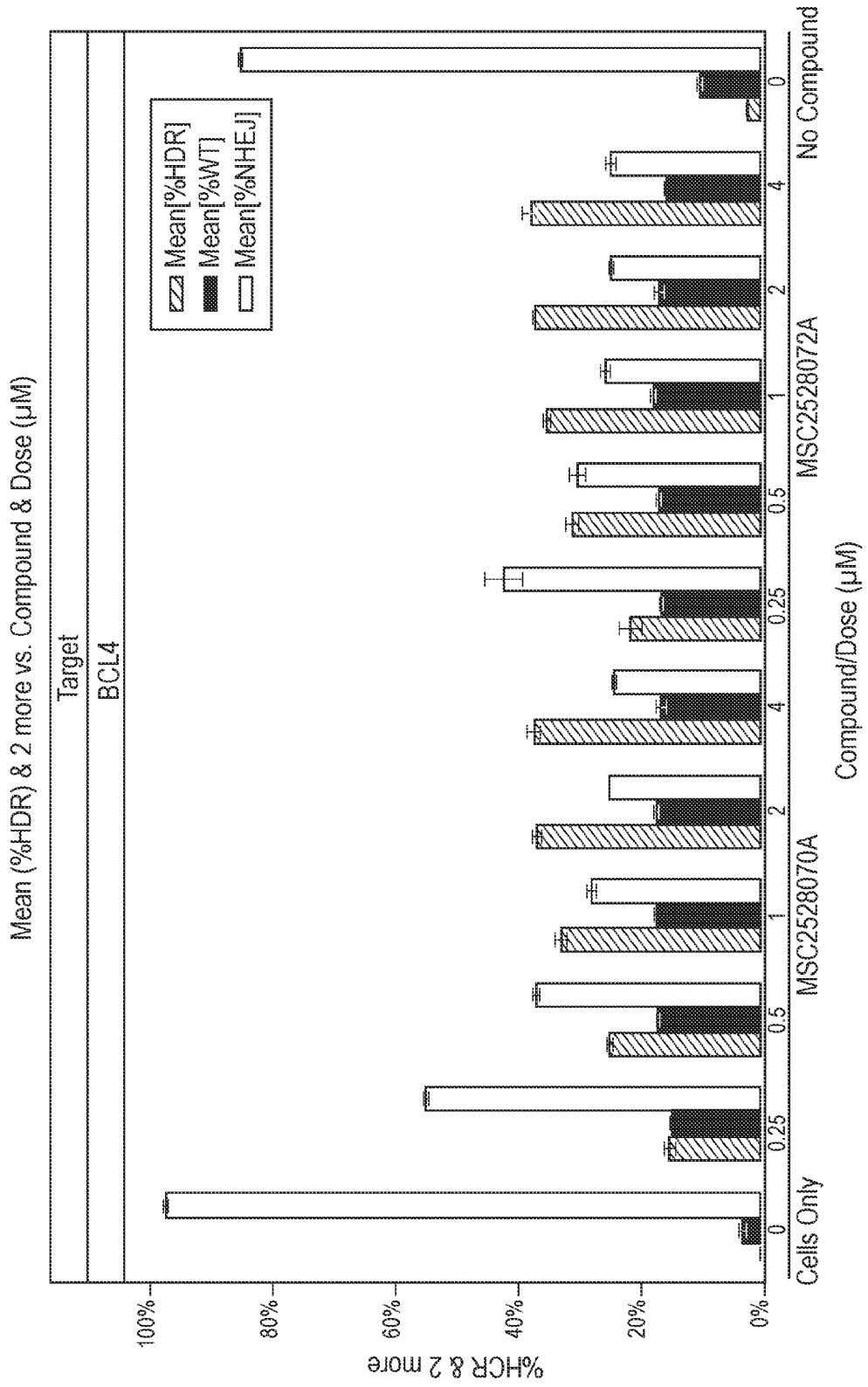

Mean (%HDR) & 2 more vs. Compound & Dose (µM)
Target
BCL4
Mean[%HDR]
Mean[%WT]
Mean[%NHEJ]
%HDR & 2 more
100%
80%
60%
40%
20%
0%
Cells Only
MSC2528070A
MSC2528072A
No Compound
0
0.25
0.5
1
2
4
0.25
0.5
1
2
4
0
Compound/Dose (µM)

FIG. 14

Target	Compound	Replicate	Dose (µM)	Wash	GLO #	%HDR	%MMEJ	%WT	%NHEJ
BCL4	MSC2528070A	1	0.25	0	320155	15.18	16.27	54.2	14.35
BCL4	MSC2528070A	1	0.25	1	353850	11.45	6.39	69.4	12.76
BCL4	MSC2528070A	1	0.5	0	326970	24.92	22.17	36.1	16.81
BCL4	MSC2528070A	1	0.5	1	347420	17.31	8.83	59.4	14.46
BCL4	MSC2528070A	1	1	0	323068	33.44	23.14	26.7	16.72
BCL4	MSC2528070A	1	1	1	351495	20.64	9.65	53.8	15.91
BCL4	MSC2528070A	1	2	0	326186	37.25	22.63	24.5	15.62
BCL4	MSC2528070A	1	2	1	345091	22.8	12.41	48.1	16.69
BCL4	MSC2528070A	1	4	0	325850	37.76	23.42	23.5	15.32
BCL4	MSC2528070A	1	4	1	349433	24.8	16.19	42.3	16.71
BCL4	MSC2528070A	2	0.25	0	338420	14.19	16.31	54.9	14.6
BCL4	MSC2528070A	2	0.25	1	347383	8.63	4.85	54	32.52
BCL4	MSC2528070A	2	0.5	0	323623	24.07	22.71	37	16.22
BCL4	MSC2528070A	2	0.5	1	353667	16.62	7.93	60.4	15.05
BCL4	MSC2528070A	2	1	0	338612	31.66	23.11	28.1	17.13
BCL4	MSC2528070A	2	1	1	346457	21.01	11.42	52	15.57

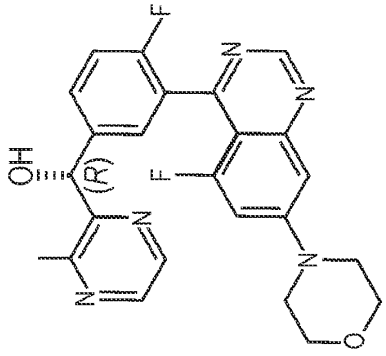
FIG. 15

BCL4	MSC2528070A	2	2	0	334296	35.48	23.56	24.6	16.36
BCL4	MSC2528070A	2	2	1	358748	23.43	13.77	46.6	16.2
BCL4	MSC2528070A	2	4	0	337583	35.73	23.31	24	16.96
BCL4	MSC2528070A	2	4	1	357500	26.61	17.73	39.2	16.46
BCL4	MSC2528072A	1	0.25	0	330852	23.05	21.63	38.9	16.42
BCL4	MSC2528072A	1	0.25	1	357754	15.1	7.64	64.1	13.16
BCL4	MSC2528072A	1	0.5	0	331349	31.56	23.18	28.4	16.86
BCL4	MSC2528072A	1	0.5	1	346549	19.98	9.14	55.1	15.78
BCL4	MSC2528072A	1	1	0	334431	35.32	22.42	24.5	17.76
BCL4	MSC2528072A	1	1	1	352045	22.39	11.55	50.1	15.96
BCL4	MSC2528072A	1	2	0	339531	36.85	22.12	23.9	17.13
BCL4	MSC2528072A	1	2	1	359373	24.95	15.43	43.6	16.02
BCL4	MSC2528072A	1	4	0	354580	38.68	22.56	23.4	15.36
BCL4	MSC2528072A	1	4	1	367775	30.9	19.15	32.4	17.55
BCL4	MSC2528072A	2	0.25	0	334415	19.08	20.13	45	15.79
BCL4	MSC2528072A	2	0.25	1	346576	13.49	7.18	63.6	15.73
BCL4	MSC2528072A	2	0.5	0	337702	29.52	23.34	31.2	15.94
BCL4	MSC2528072A	2	0.5	1	342036	19.53	8.93	55.7	15.84

FIG. 15 CONT.

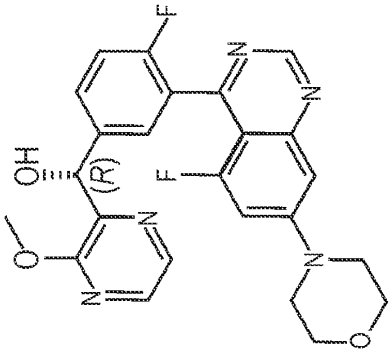
BCL4	MSC2528072A	2	1	0	340510	34.18	23.09	25.9	16.83
BCL4	MSC2528072A	2	1	1	354407	20.54	10.4	54	15.06
BCL4	MSC2528072A	2	2	0	344707	37.05	22.84	24.5	15.61
BCL4	MSC2528072A	2	2	1	358515	24.41	14.61	44.7	16.28
BCL4	MSC2528072A	2	4	0	343653	36.31	22.77	25.3	15.62
BCL4	MSC2528072A	2	4	1	352241	30.01	20.65	33.5	15.84
BCL4	Cells Only	2	0	1	399360	0	0	97.5	2.5
BCL4	Cells Only	1	0	1	443125	0	0	96.8	3.2
BCL4	Cells Only	2	0	0	280599	0	0	97.3	2.7
BCL4	Cells Only	1	0	0	288637	0	0	96.8	3.2
BCL4	No Compound	2	0	1	345927	1.95	2.76	83.8	11.49
BCL4	No Compound	1	0	1	369153	2.17	2.87	84.5	10.46
BCL4	No Compound	2	0	0	331744	1.96	3.15	84.8	10.09
BCL4	No Compound	1	0	0	329112	2.12	3.46	85.2	9.22

FIG. 15 CONT.



MSC2528072A

(R)-[4-Fluoro-3-(5-fluoro-7-morpholin-4-yl-quinazolin-4-yl)-phenyl]-(3-methyl-pyrazin-2-yl]-methanol



MSC2528070A

(R)-[4-Fluoro-3-(5-fluoro-7-morpholin-4-yl-quinazolin-4-yl)-phenyl]-(3-methoxy-pyrazin-2-yl]-methanol

FIG. 16