FIG. 1A

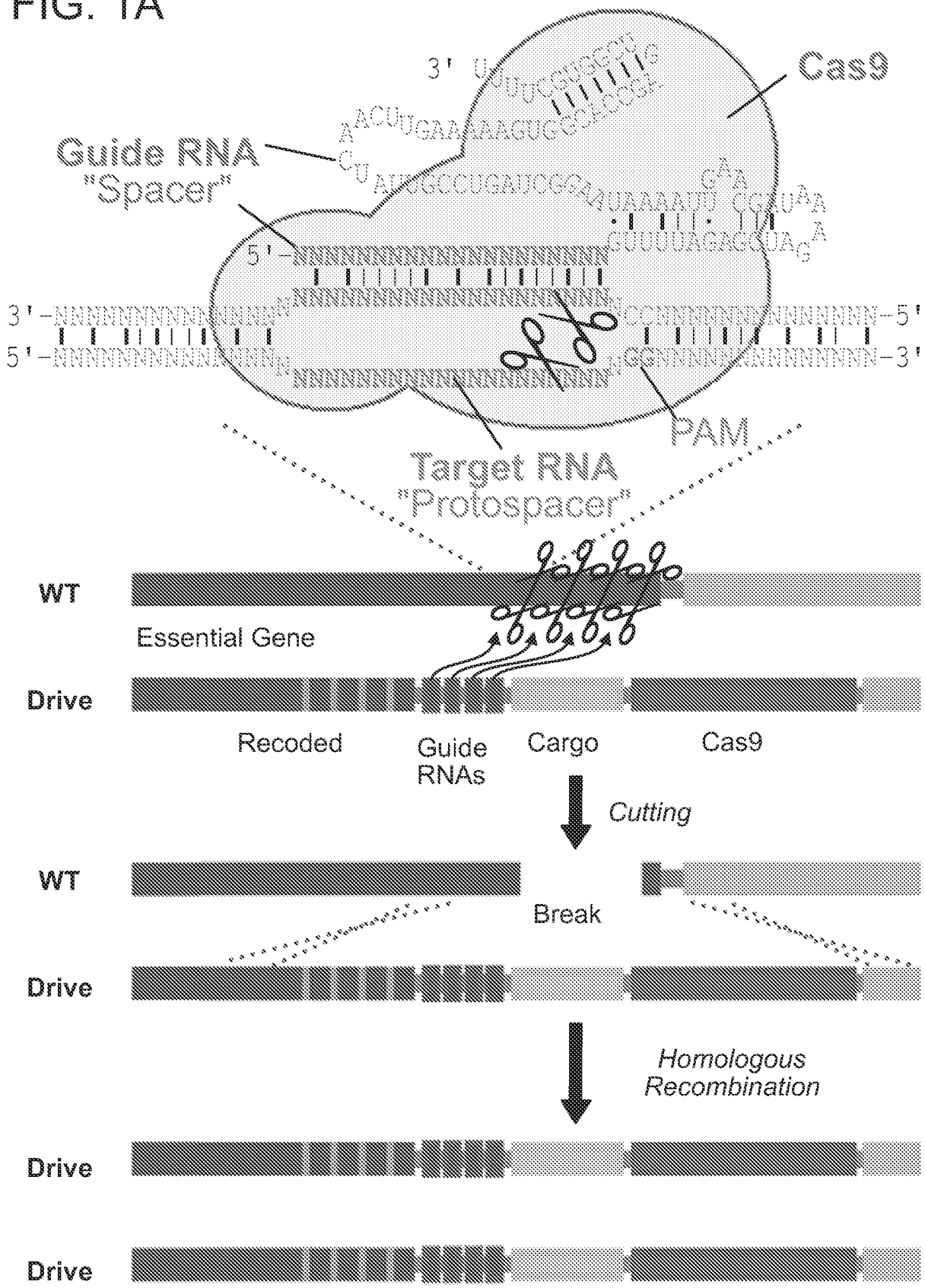

Cas9
Guide RNA
"Spacer"
Target RNA
"Protospacer"
PAM
WT
Essential Gene
Drive
Recoded
Guide RNAs
Cargo
Cas9
Cutting
WT
Break
Drive
Homologous Recombination
Drive
Drive

FIG. 1B

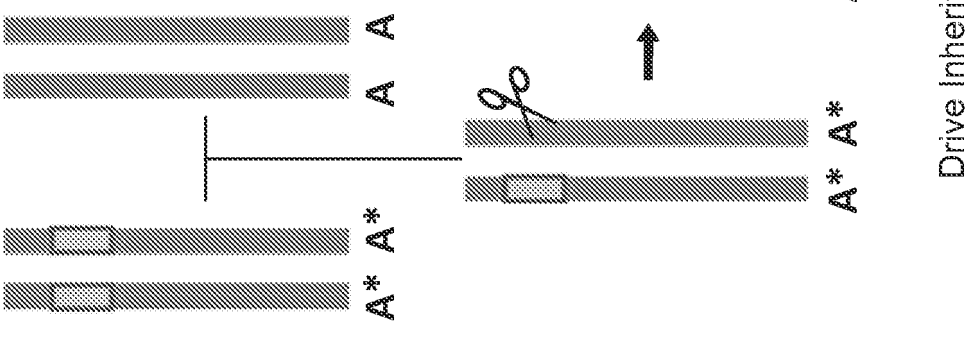


FIG. 1C

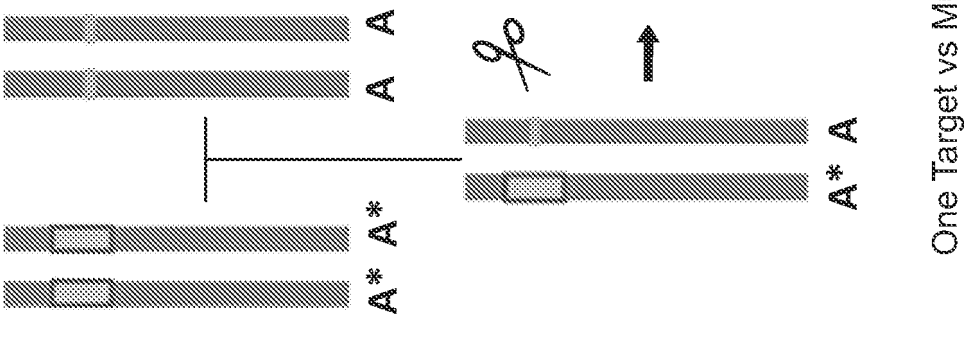


FIG. 1D

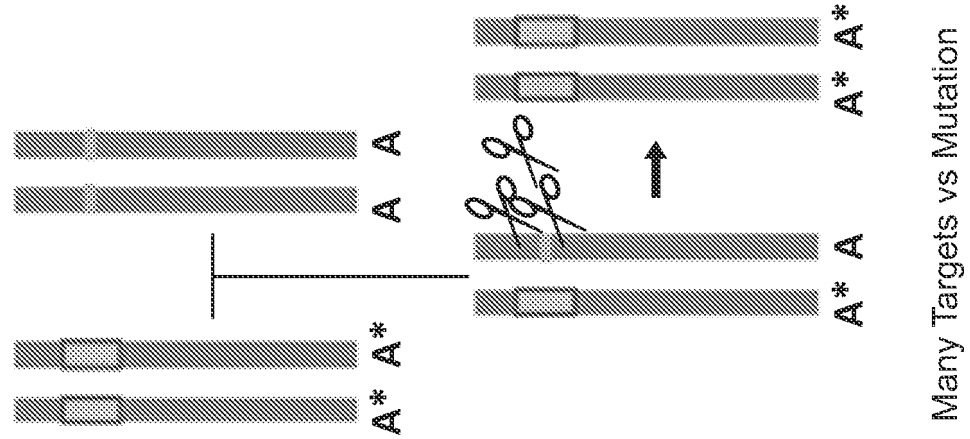


FIG. 1E

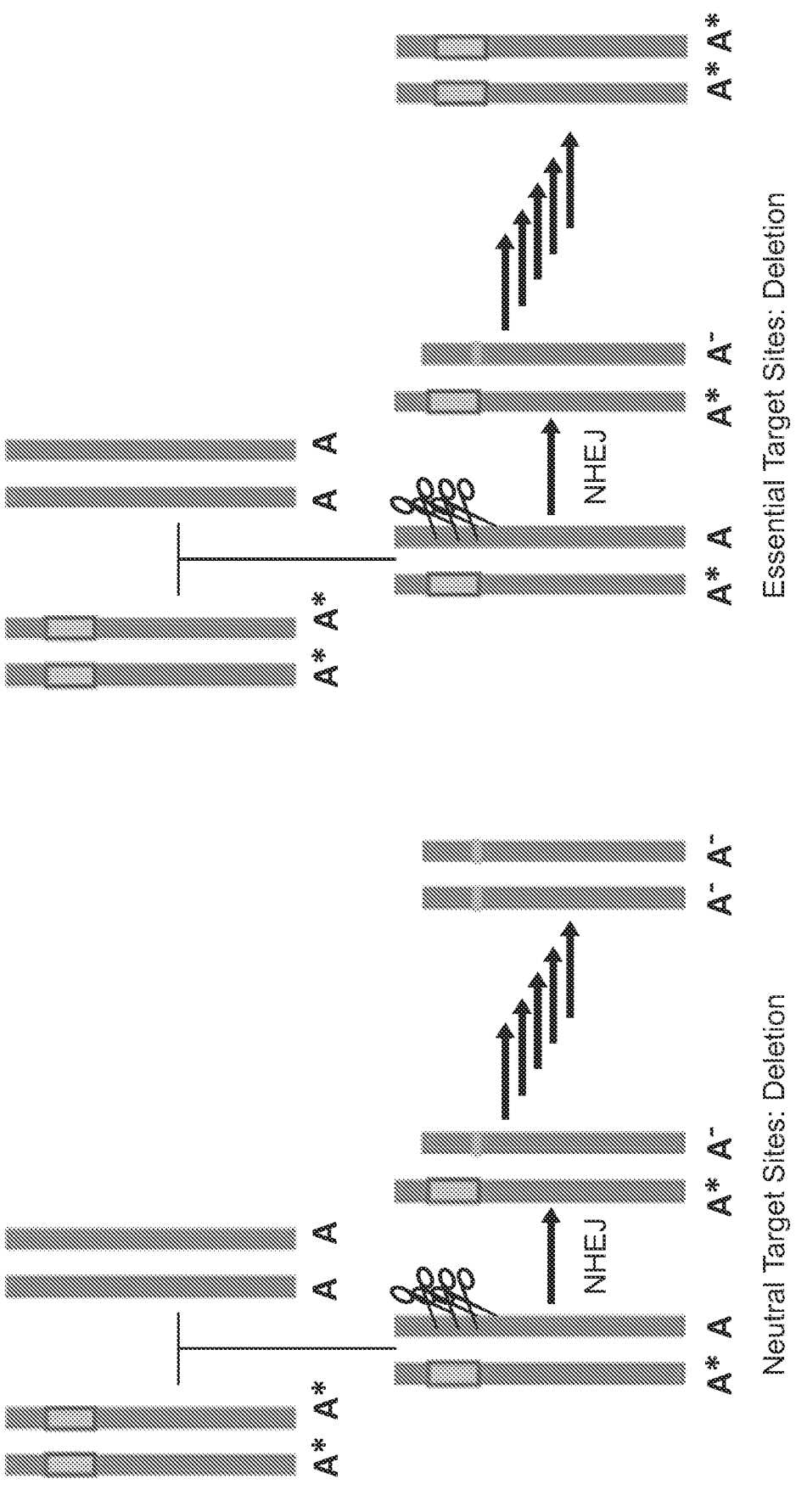

A* A*
A A
NHEJ
A* A
A* A⁻
A⁻ A⁻
Neutral Target Sites: Deletion

FIG. 1F

A* A*
A A
NHEJ
A* A
A* A⁻
A⁻ A⁻
Essential Target Sites: Deletion

FIG. 2A

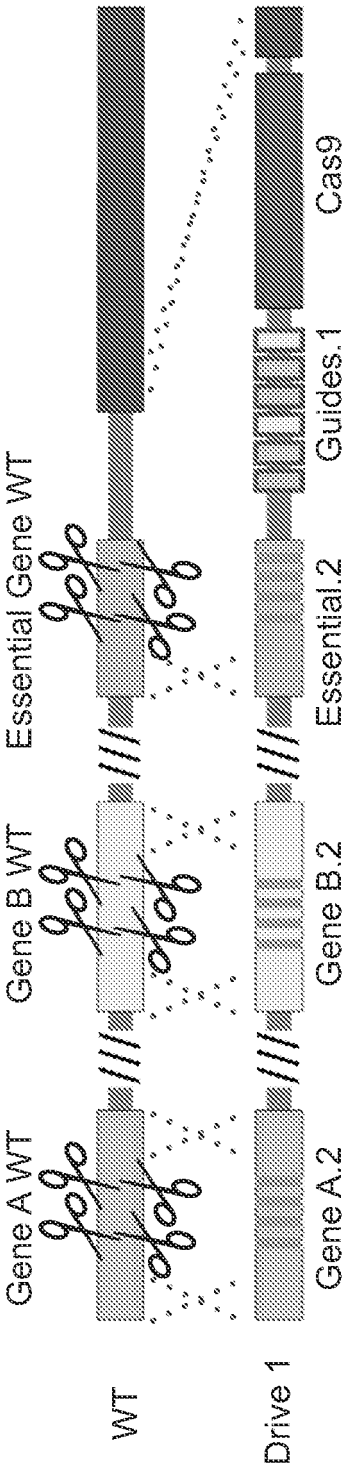


FIG. 2B

FIG. 2C

FIG. 3

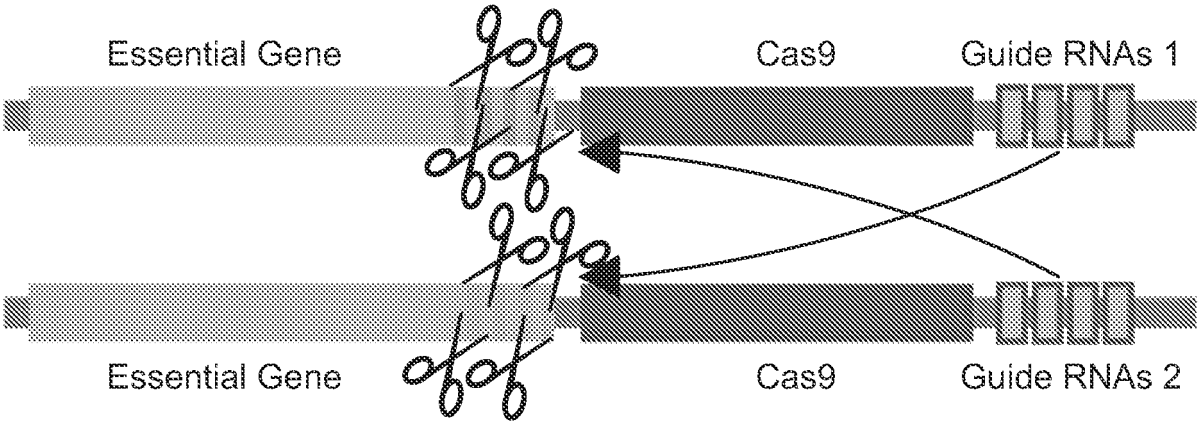

Essential Gene
Cas9
Guide RNAs 1
Essential Gene
Cas9
Guide RNAs 2

FIG. 4A

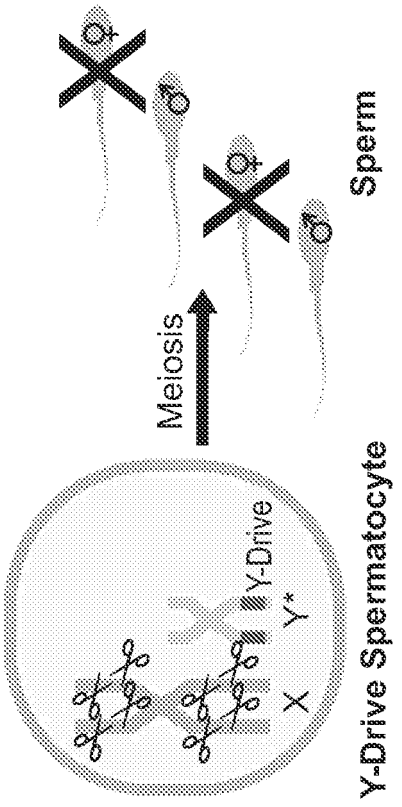


FIG.4B

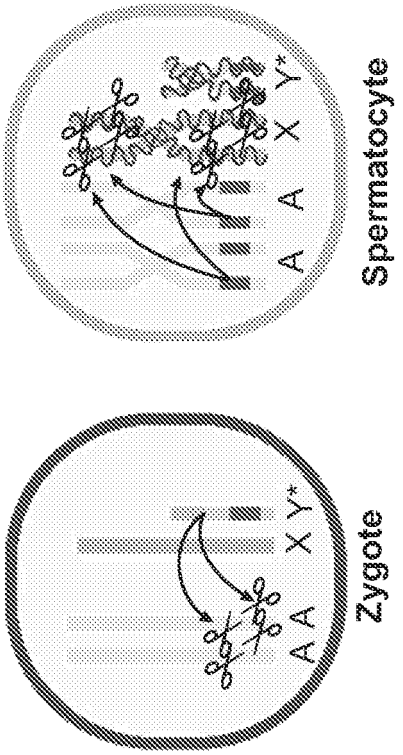
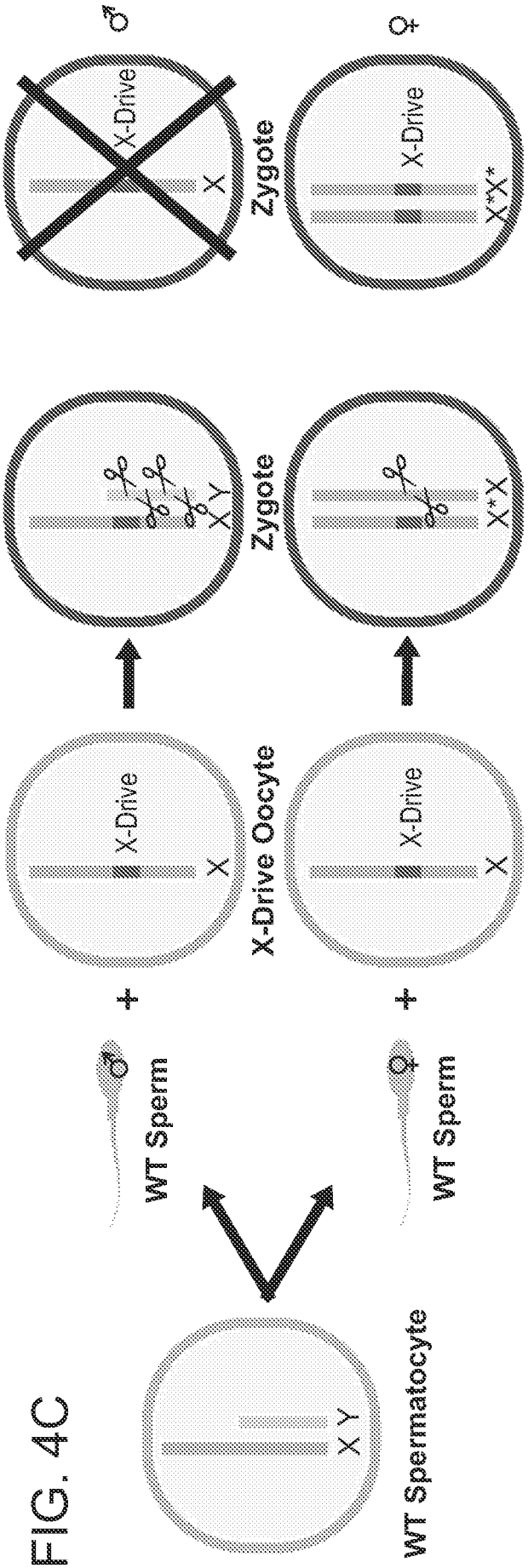


FIG. 4C



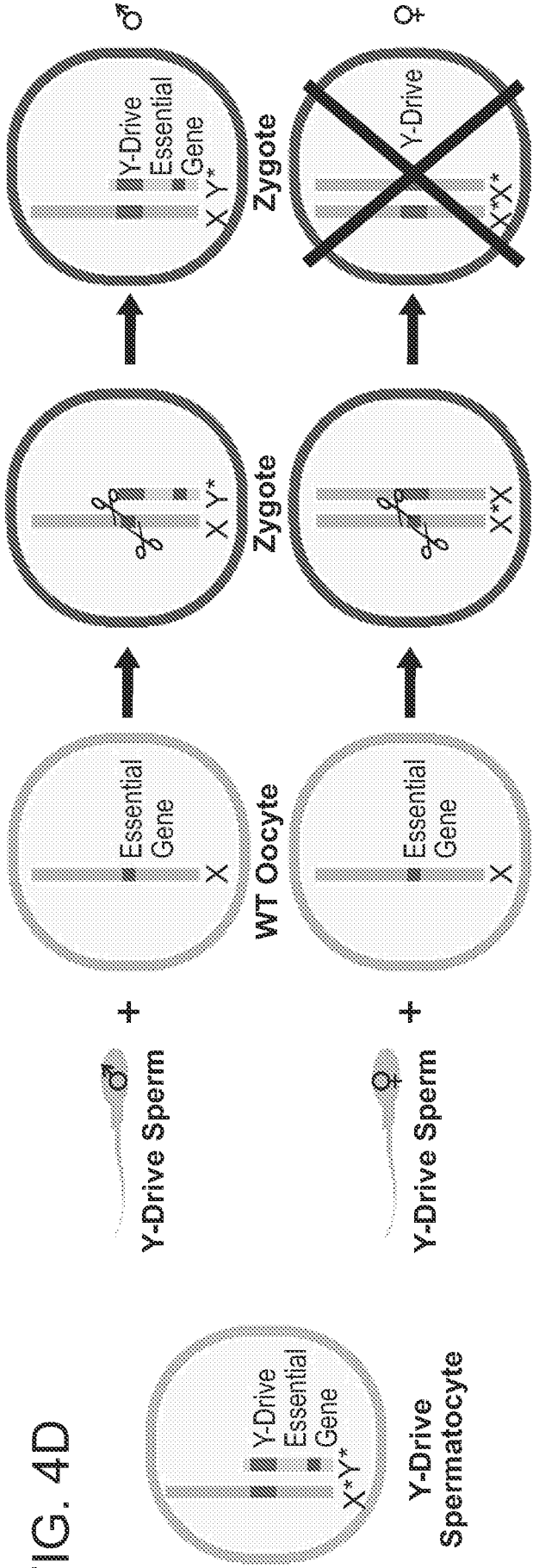


FIG. 4D

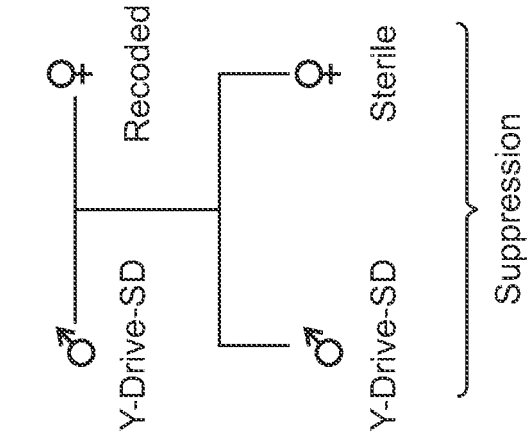


FIG. 5B

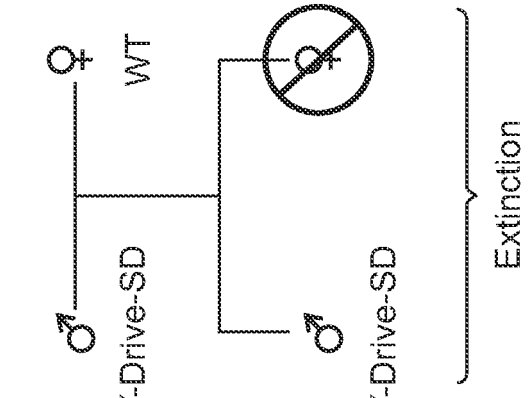


FIG. 5A

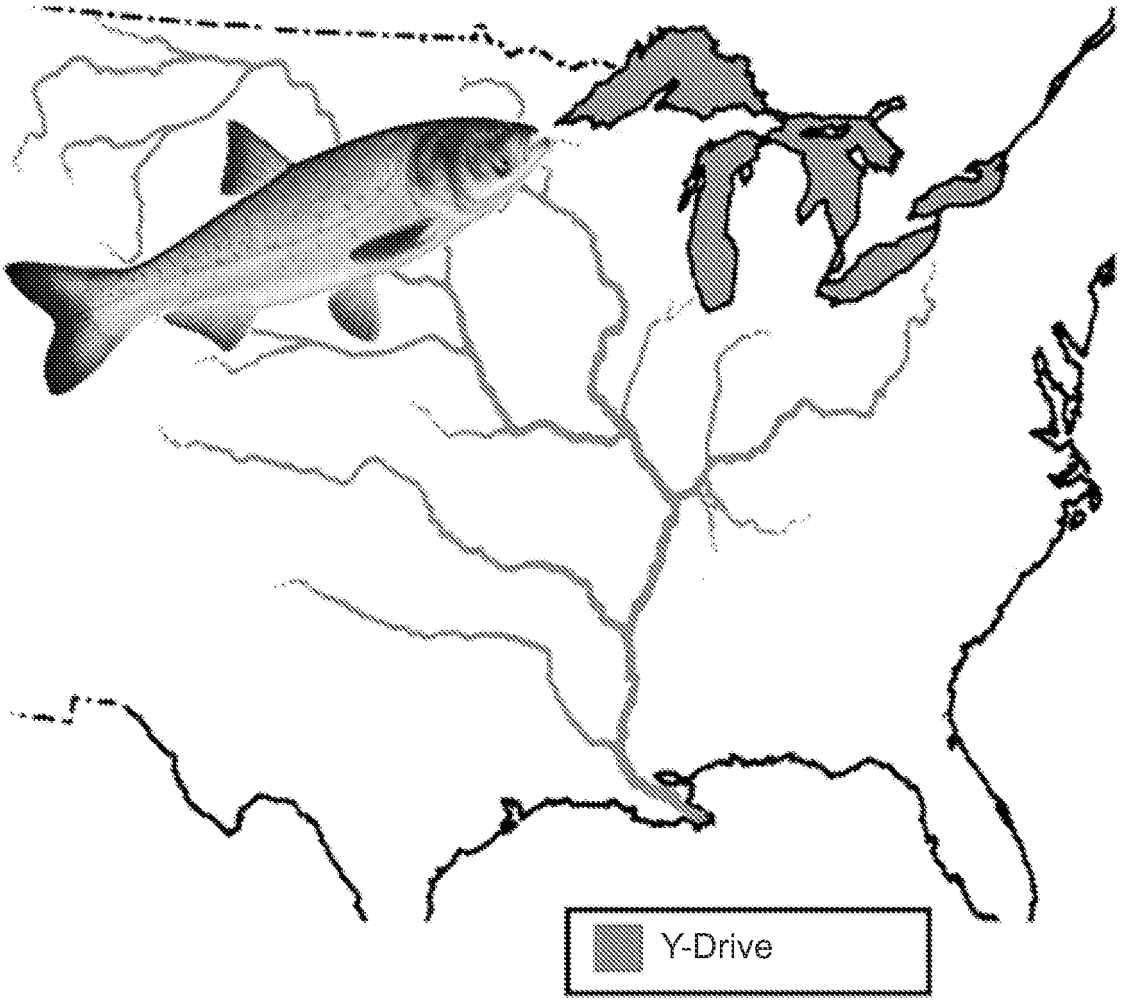


FIG. 6A

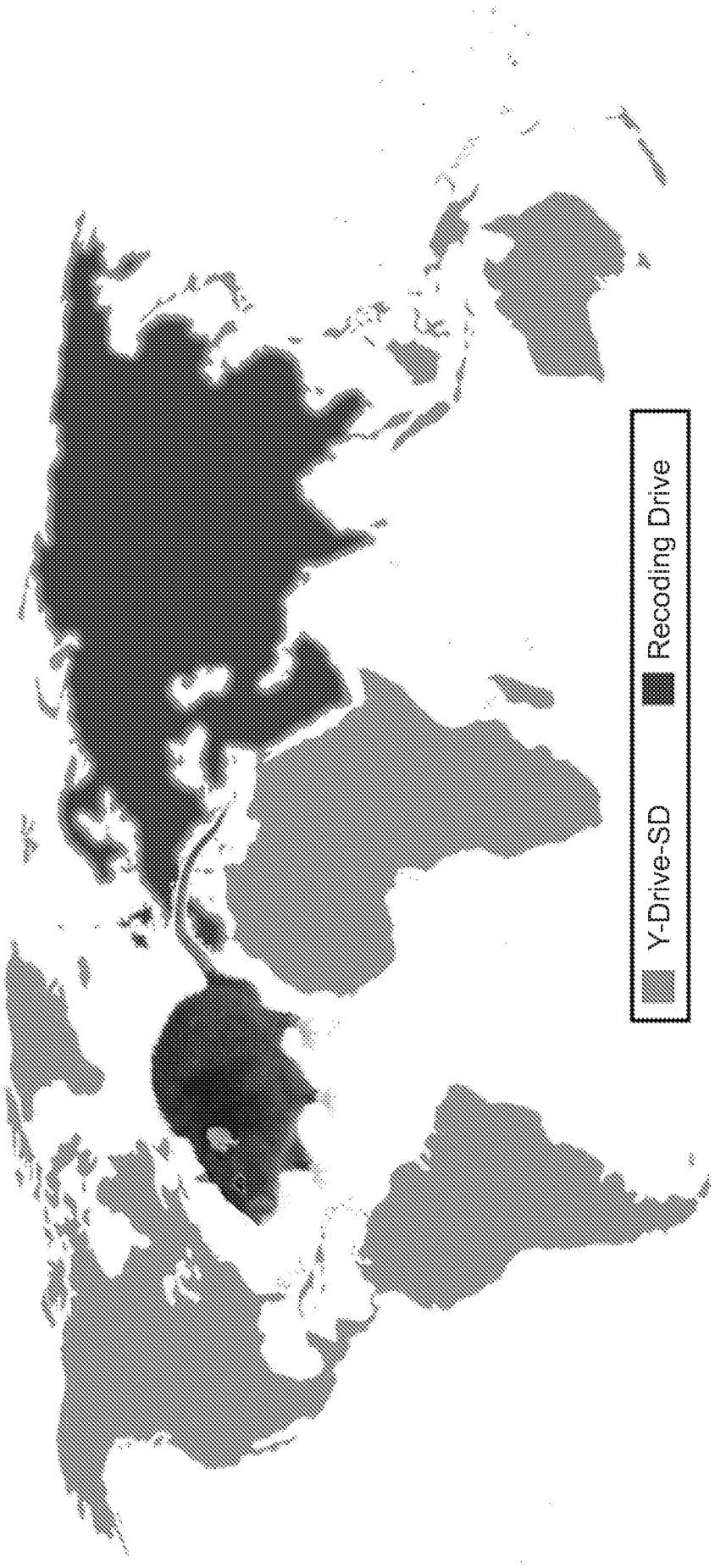


FIG.6B

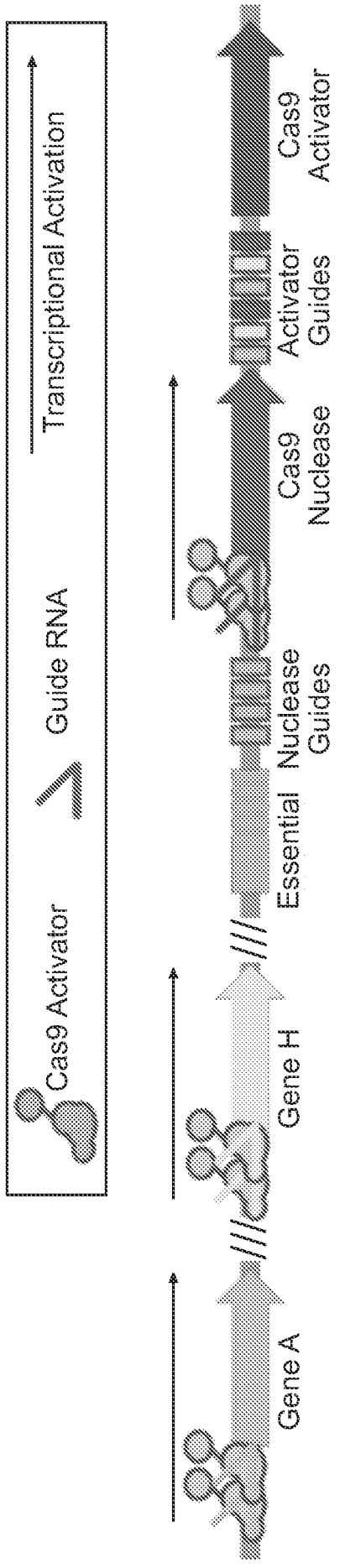

Transcriptional Activation
Guide RNA
Cas9 Activator
Gene A
Gene H
Essential Nuclease Guides
Cas9 Nuclease
Activator Guides
Cas9 Activator

FIG. 7

FIG. 8A

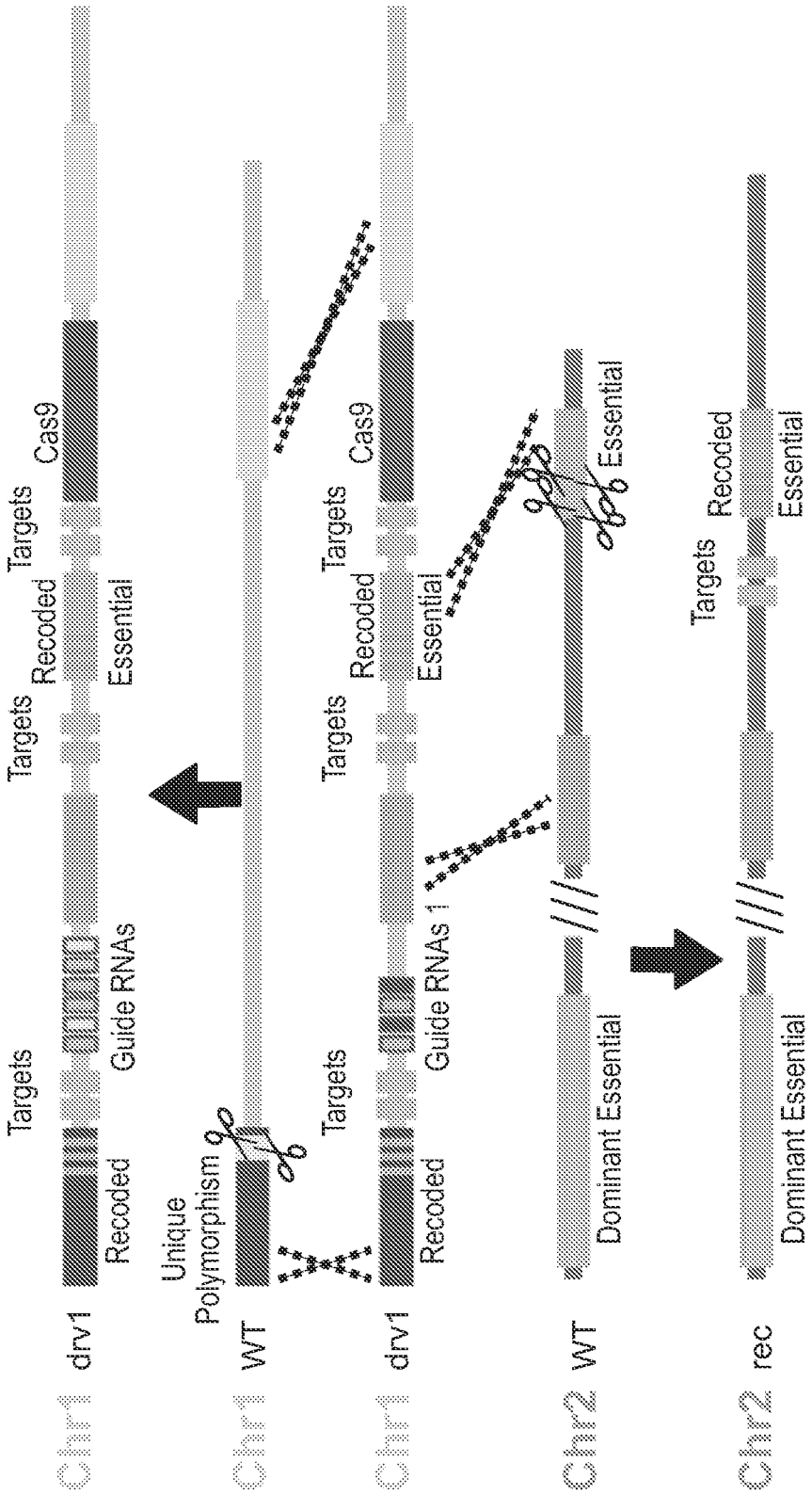

Chr1 drv1
Targets
Recoded
Guide RNAs
Targets
Recoded
Essential
Targets
Cas9
Chr1 WT
Unique Polymorphism
Targets
Chr1 drv1
Targets
Recoded
Guide RNAs 1
Targets
Recoded
Essential
Targets
Cas9
Chr2 WT
Dominant Essential
Essential
Chr2 rec
Targets
Recoded
Essential
Dominant Essential

FIG. 8B

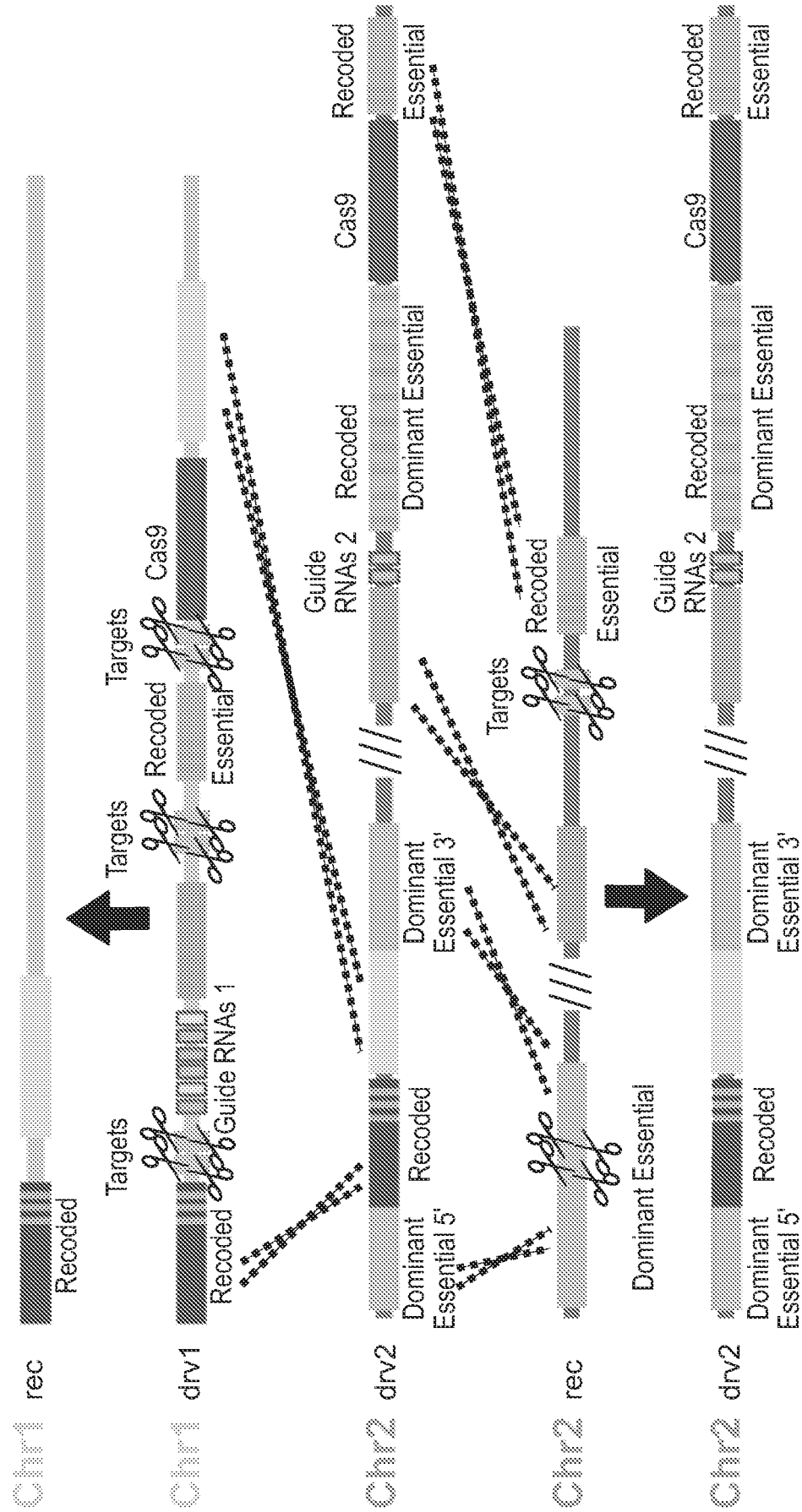

Chr1 rec
Recoded
Chr1 drv1
Targets
Recoded
Guide RNAs 1
Targets
Recoded
Cas9
Essential
Chr2 drv2
Dominant Essential 5'
Recoded
Dominant Essential 3'
Guide RNAs 2
Recoded
Cas9
Dominant Essential
Chr2 rec
Dominant Essential
Targets
Recoded
Essential
Chr2 drv2
Dominant Essential 5'
Recoded
Dominant Essential 3'
Guide RNAs 2
Recoded
Cas9
Dominant Essential

FIG. 8C

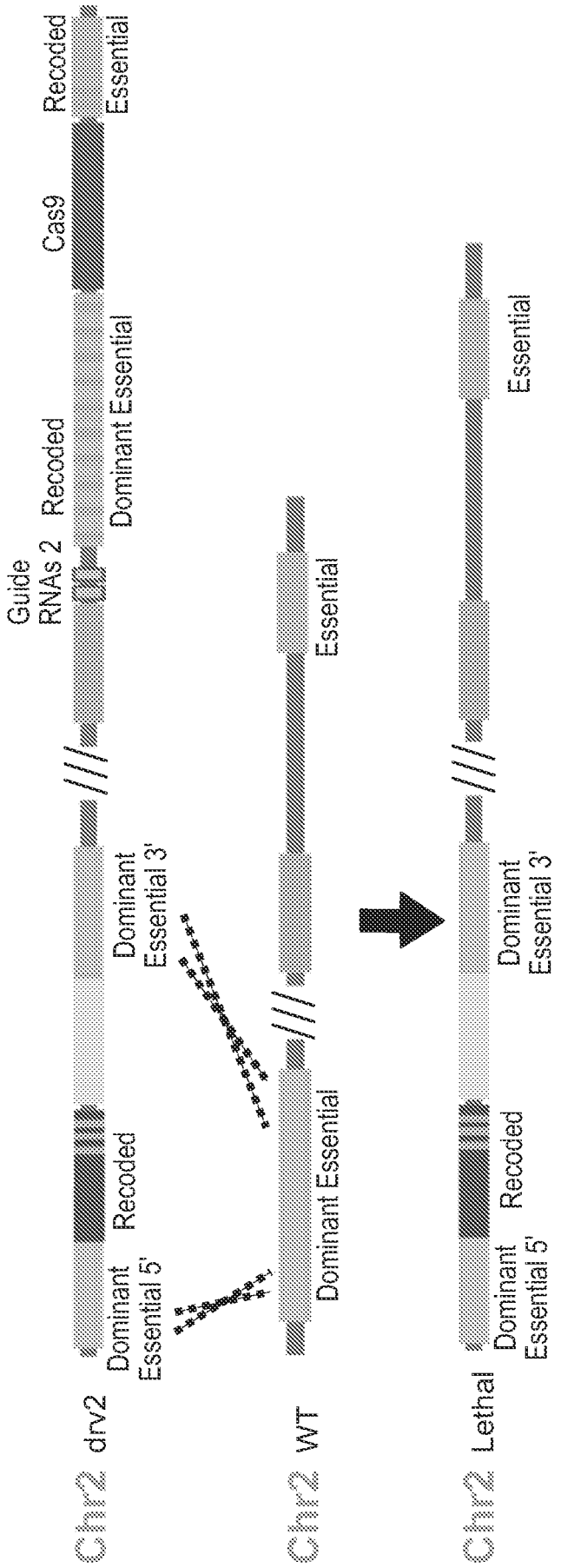
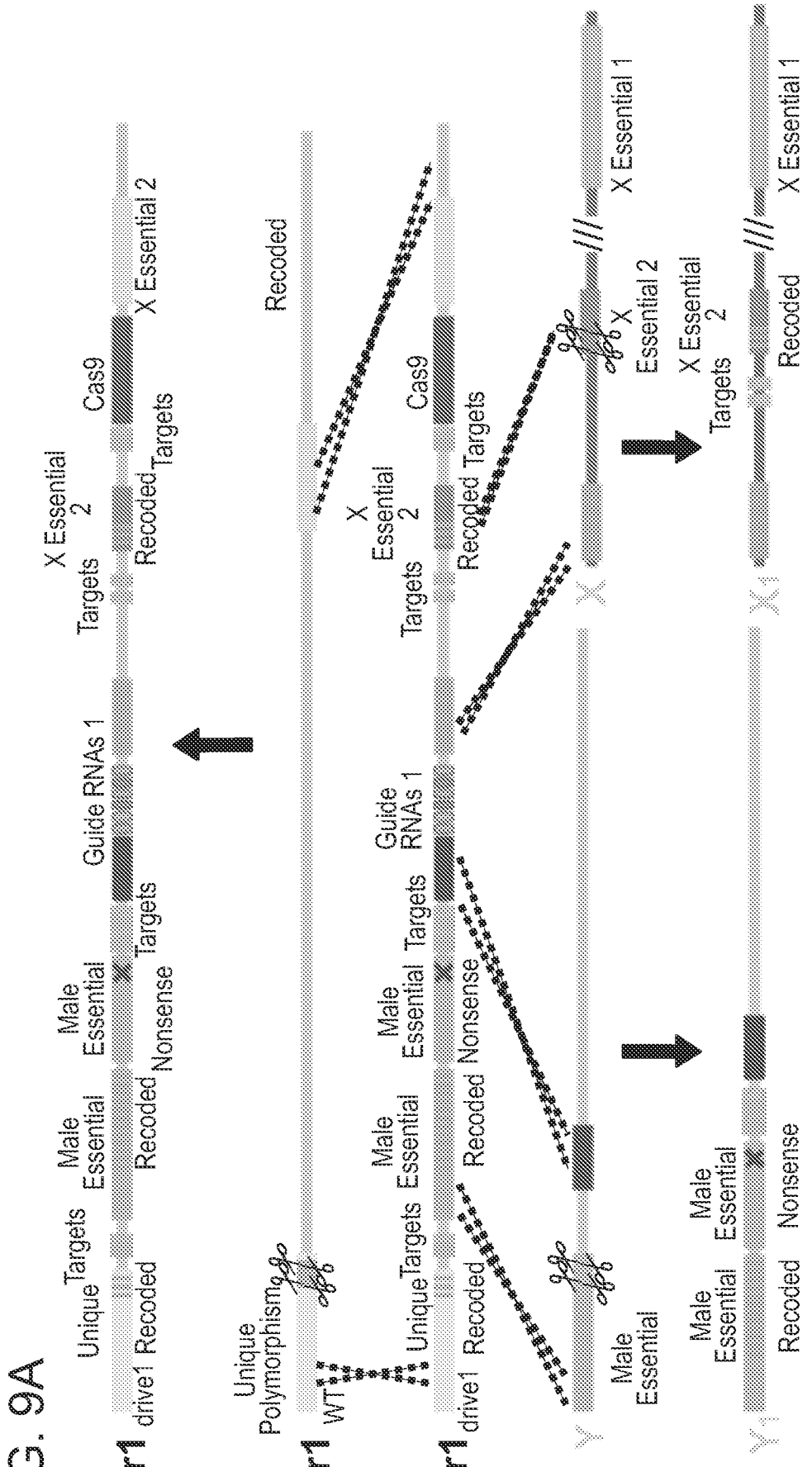

Chr2 drv2
Dominant Essential 5'
Recoded
Dominant Essential 3'
Guide RNAs 2
Recoded
Cas9
Recoded
Dominant Essential
Essential
Chr2 WT
Dominant Essential
Essential
Chr2 Lethal
Dominant Essential 5'
Recoded
Dominant Essential 3'
Essential

FIG. 9A


Chr1
drive1
Recoded
Unique Targets
Male Essential
Recoded
Nonsense
Male Essential Targets
Guide RNAs 1
Cas9
X Essential 2 Targets
X Essential 2
Recoded
Unique Polymorphism
WT
Unique Targets
Male Essential
Male Essential
Recoded
Nonsense
Male Essential Targets
Guide RNAs 1
Cas9
X Essential 2 Targets
X Essential 2
Recoded
X Essential 1
Male Essential
Male Essential
Recoded
Nonsense
X Essential 2 Targets
X Essential 2
Recoded
X Essential 1
Y
Y1
X
X1

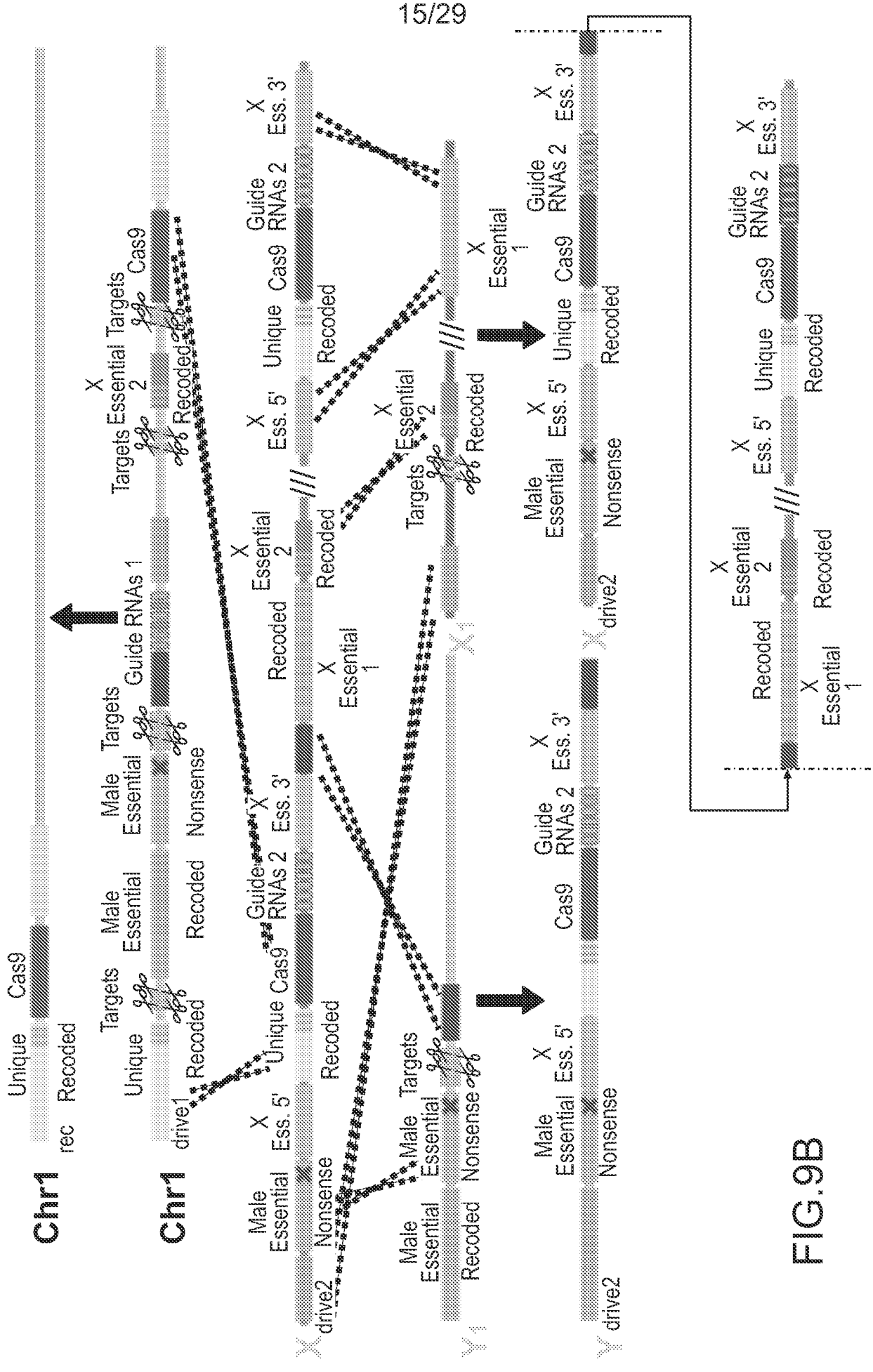

Chr1
rec
Unique
Cas9
Recoded
Chr1
drive1
Unique
Targets
Male
Essential
Targets
Recoded
Nonsense
Male
Essential
X
Ess. 5'
Unique
Cas9
Guide
RNAs 2
X
Ess. 3'
Recoded
X
Essential
1
X
Essential
2
Guide
RNAs 1
Male
Essential
Nonsense
Recoded
drive2
Y1
Male
Essential
Targets
Recoded
Nonsense
Male
Essential
X
Ess. 5'
Unique
Cas9
Guide
RNAs 2
X
Ess. 3'
Recoded
X
Essential
1
X
Essential
2
Male
Essential
Nonsense
drive2
Y
Recoded
X
Essential
2
X
Ess. 5'
Unique
Cas9
Guide
RNAs 2
X
Ess. 3'
Recoded
X
Essential
1

FIG.9B

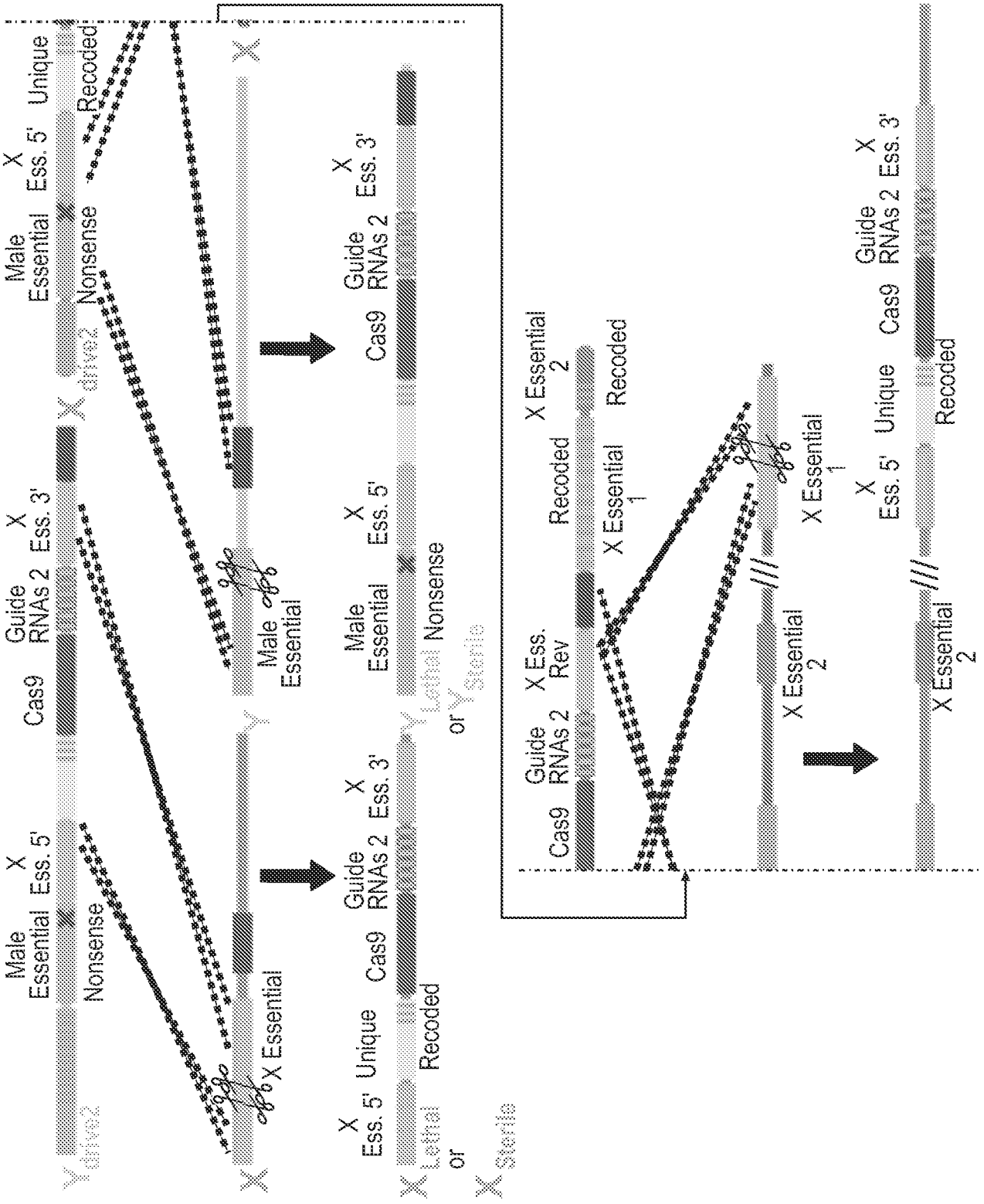

Male Essential
X Ess. 5'
Nonsense
Cas9
Guide RNAs 2
X Ess. 3'
X Essential
Unique
Recoded
Lethal or Sterile
X Ess. Rev
Essential 2
Essential 1

FIG. 9C

FIG. 10A



FIG. 10B



FIG. 10C

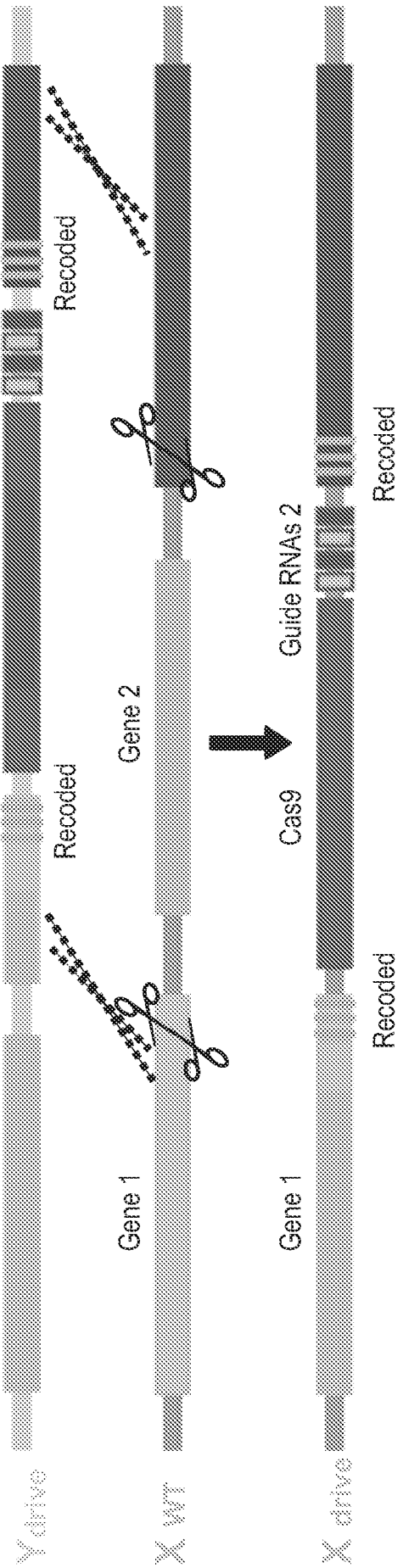


FIG. 10D

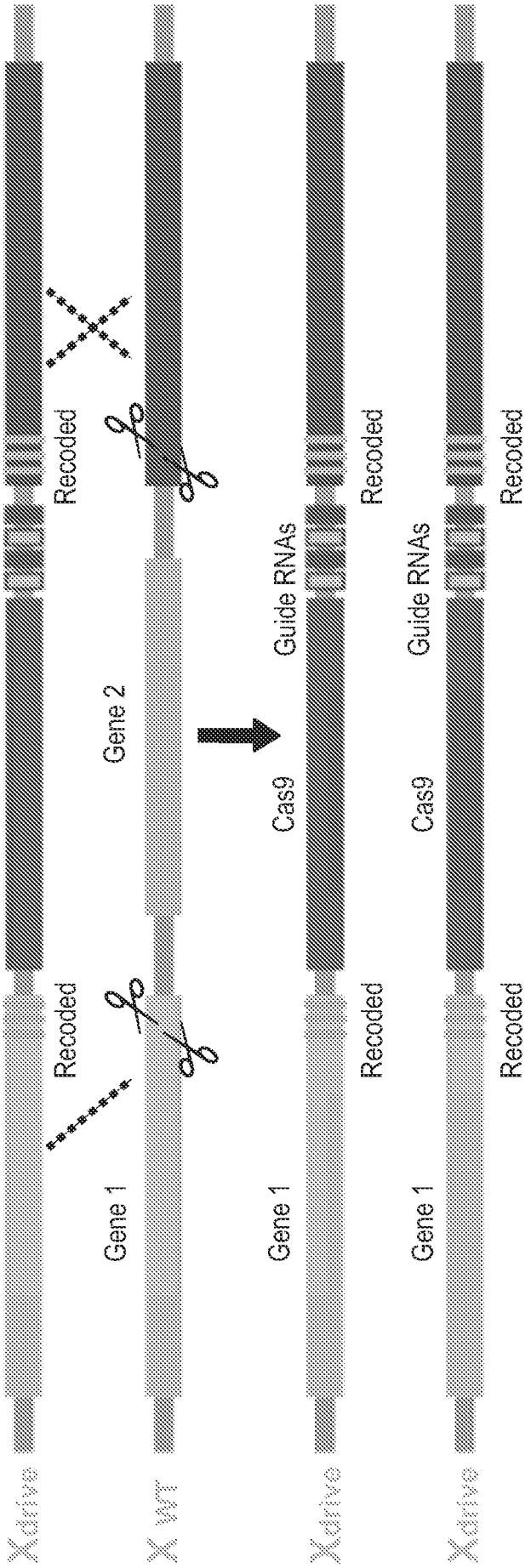

Xdrive
Cas9
Guide RNAs
Recoded
Gene 1
X WT
Gene 2
Recoded
Gene 1
Xdrive
Cas9
Guide RNAs
Recoded
Gene 1
Xdrive
Cas9
Guide RNAs
Recoded
Gene 1

FIG. 11A

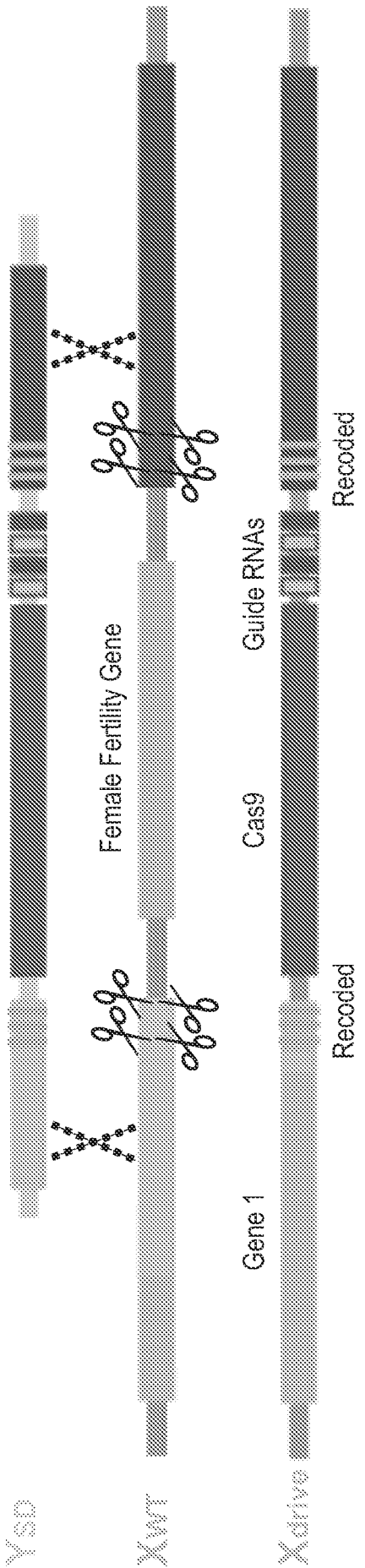


FIG. 11B

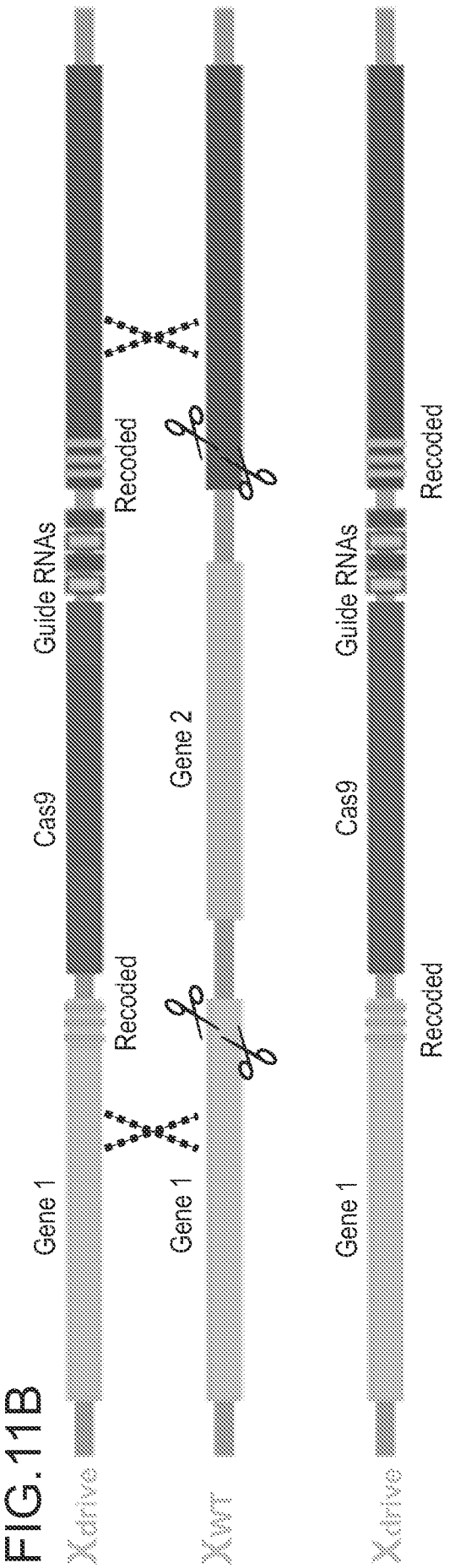


FIG. 12A

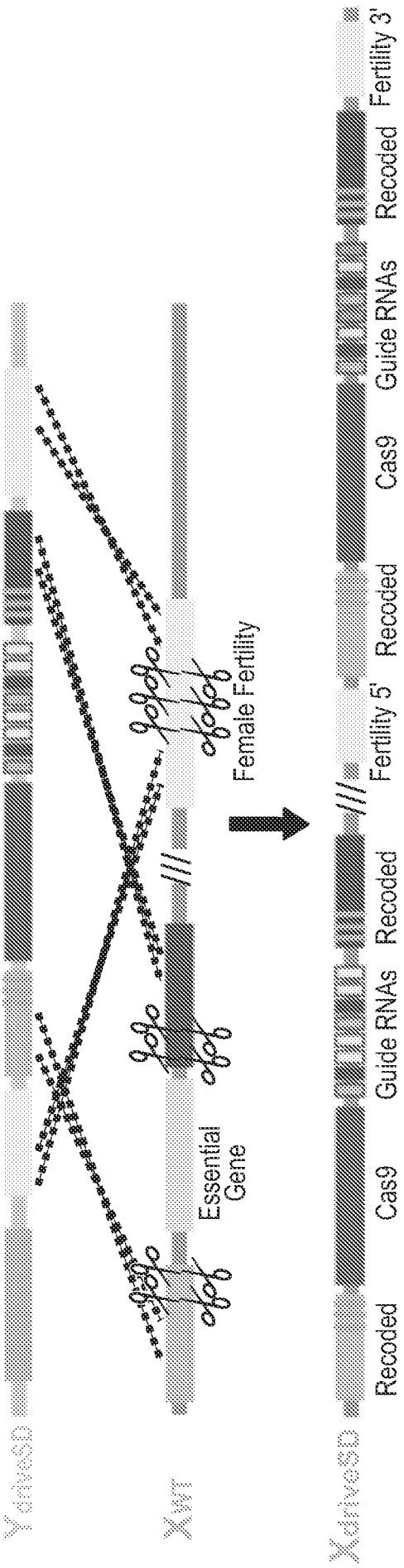


FIG. 12B

FIG. 12C

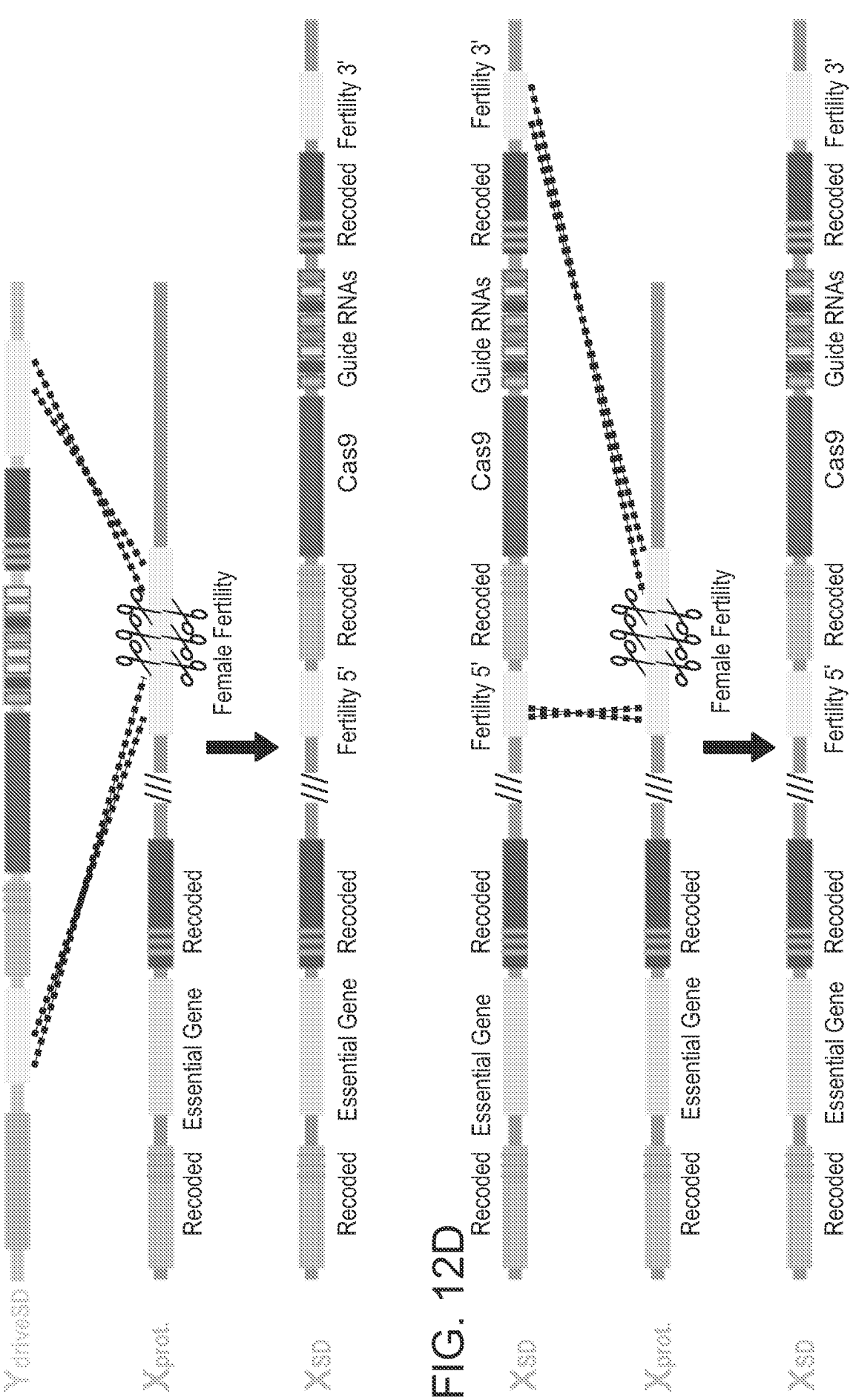
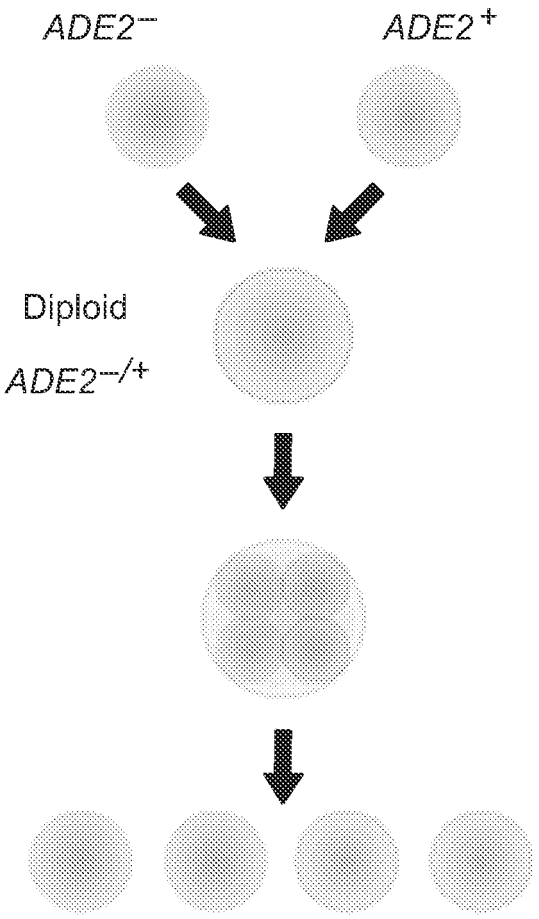


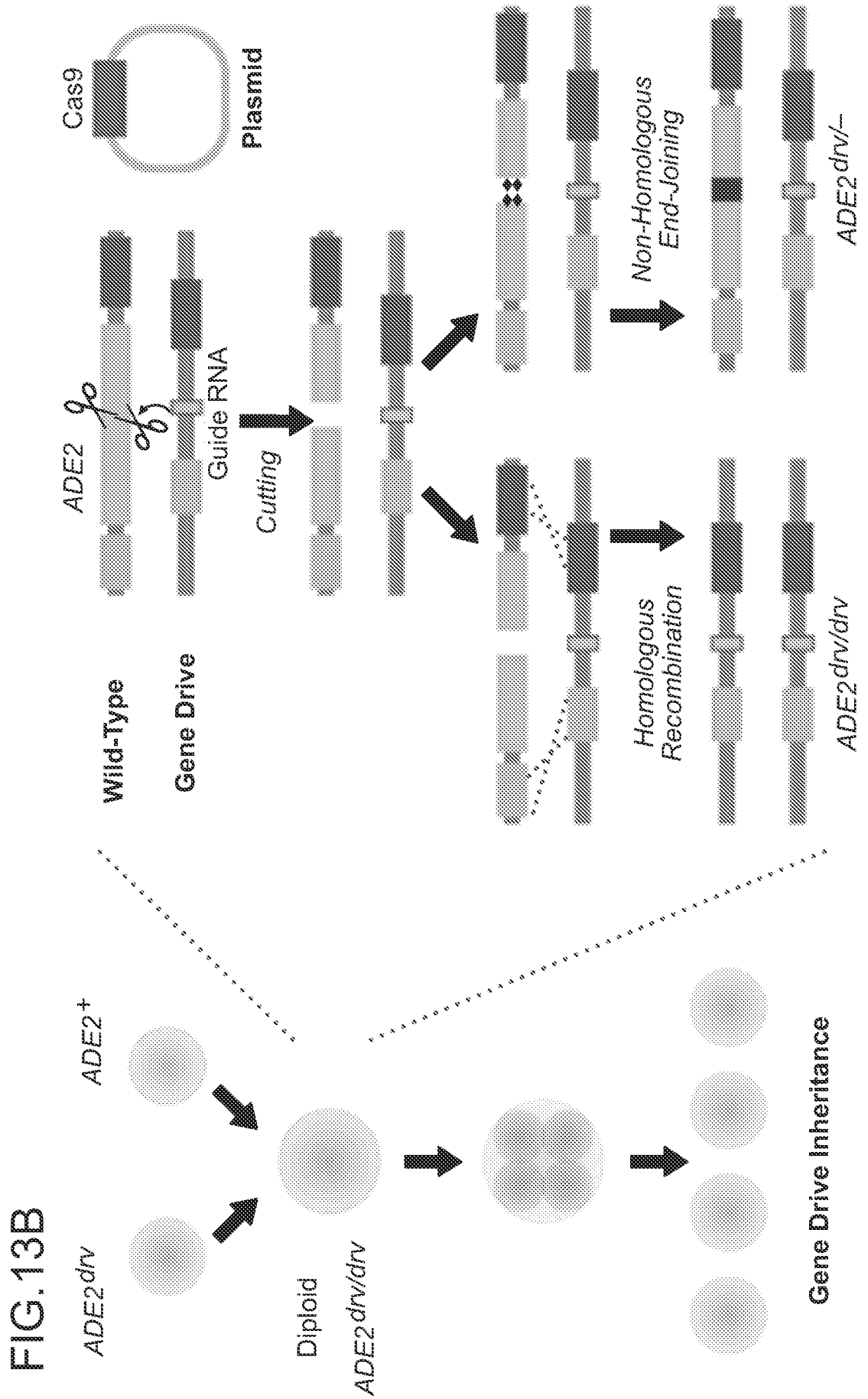
FIG. 12D

FIG. 13A



Normal Inheritance

FIG. 13B


Cas9
Plasmid
ADE2
Guide RNA
Cutting
Wild-Type
Gene Drive
Non-Homologous End-Joining
Homologous Recombination
ADE2drv/–
ADE2drv/drv
ADE2drv
ADE2+
Diploid
ADE2drv/drv
Gene Drive Inheritance

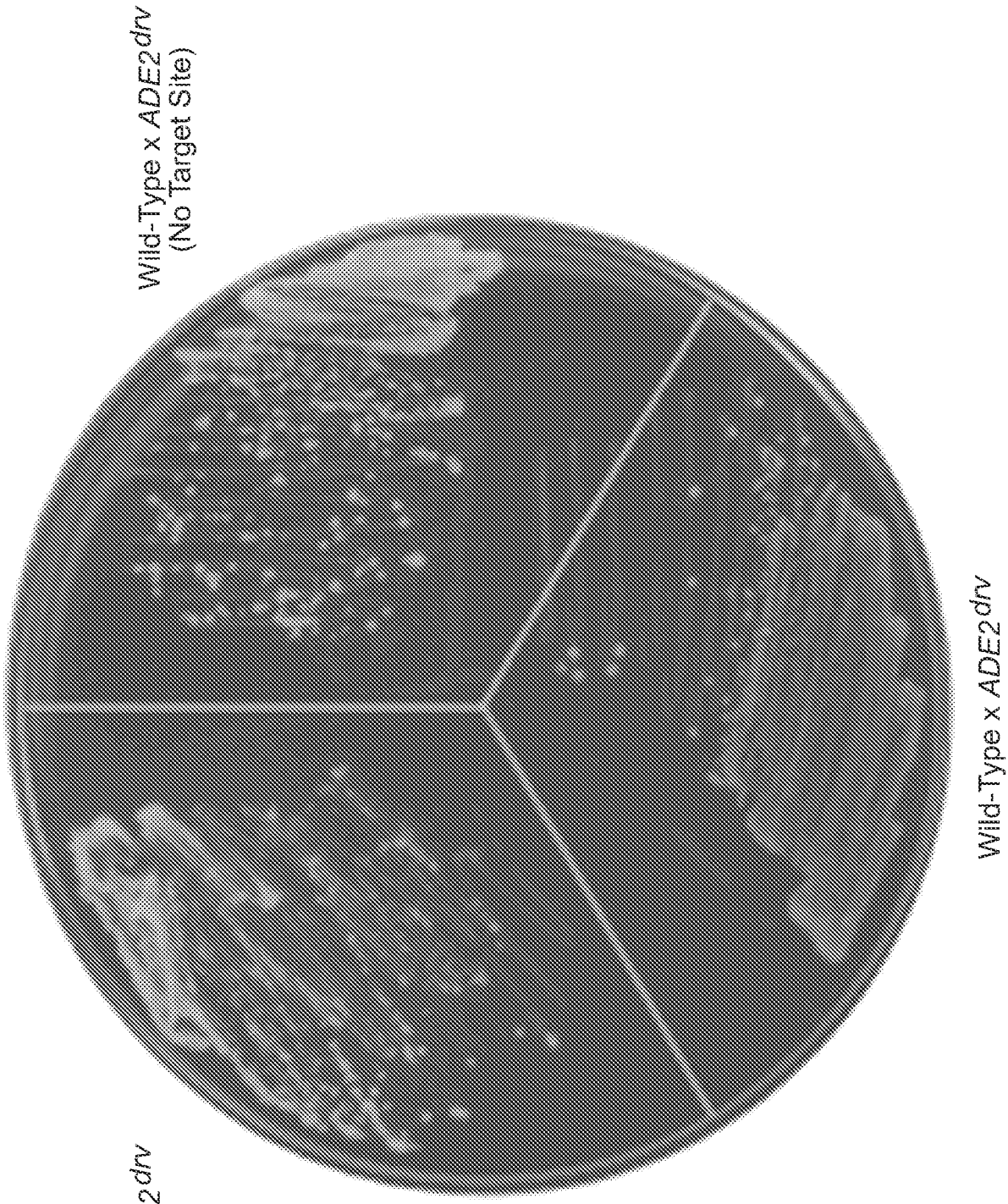

Wild-Type x ADE2drv
(No Target Site)
Wild-Type x ADE2drv
Wild-Type x ADE2drv
(No Cas9)

FIG. 13C

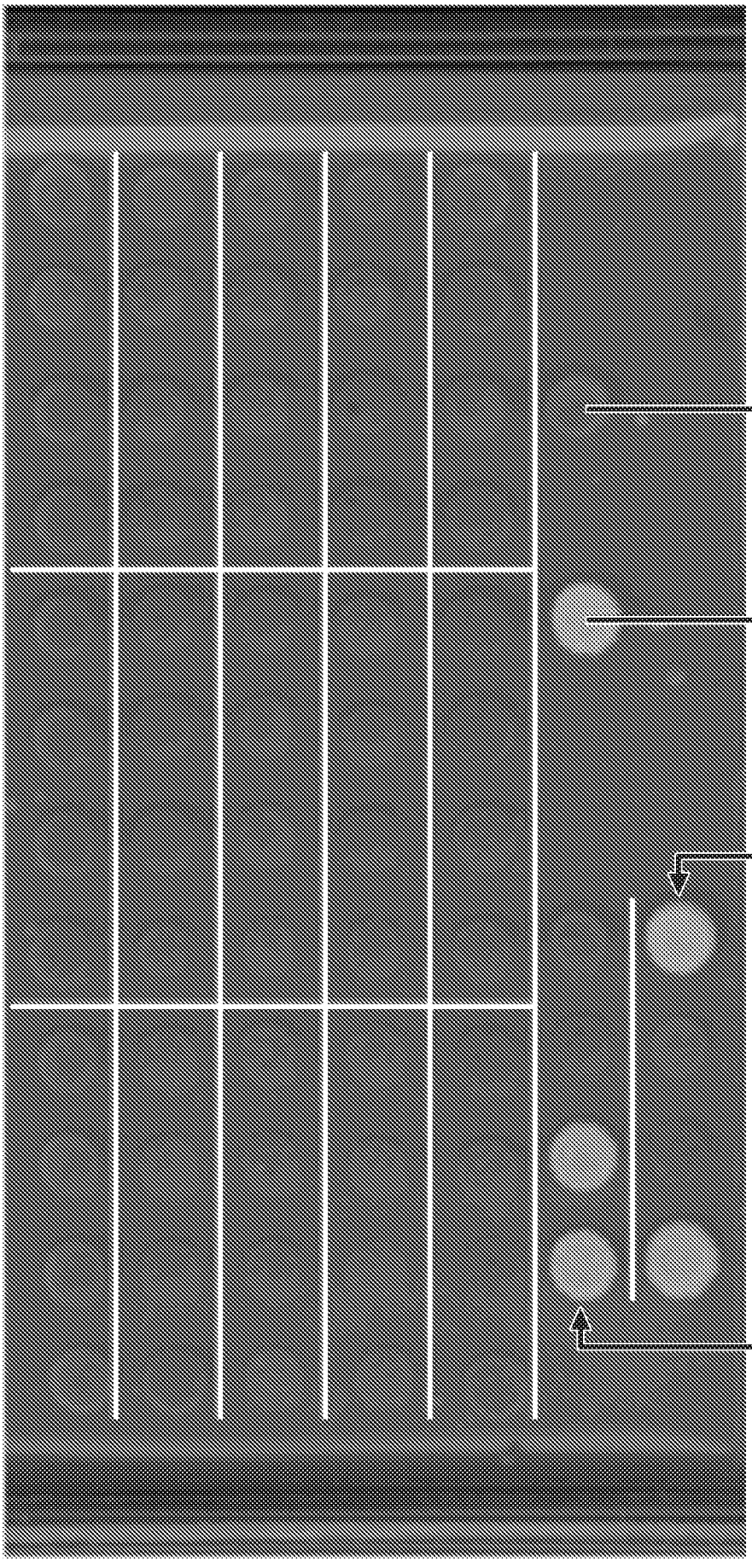


FIG. 13D

FIG. 14A

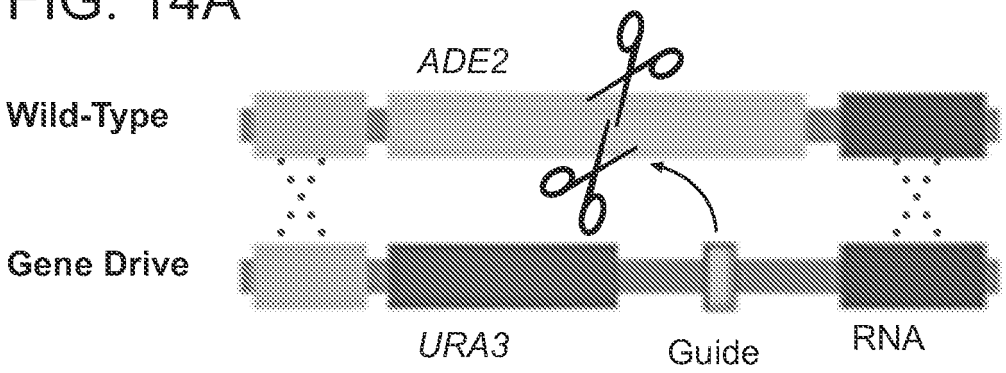


FIG. 14C

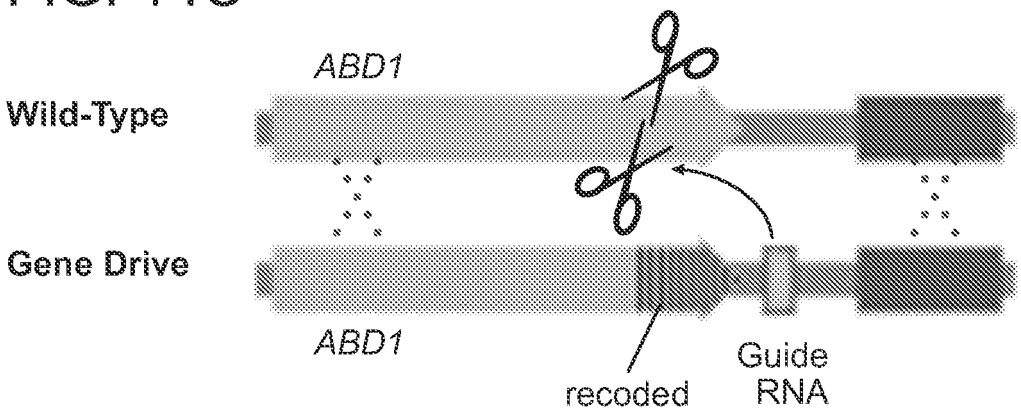


FIG. 14B

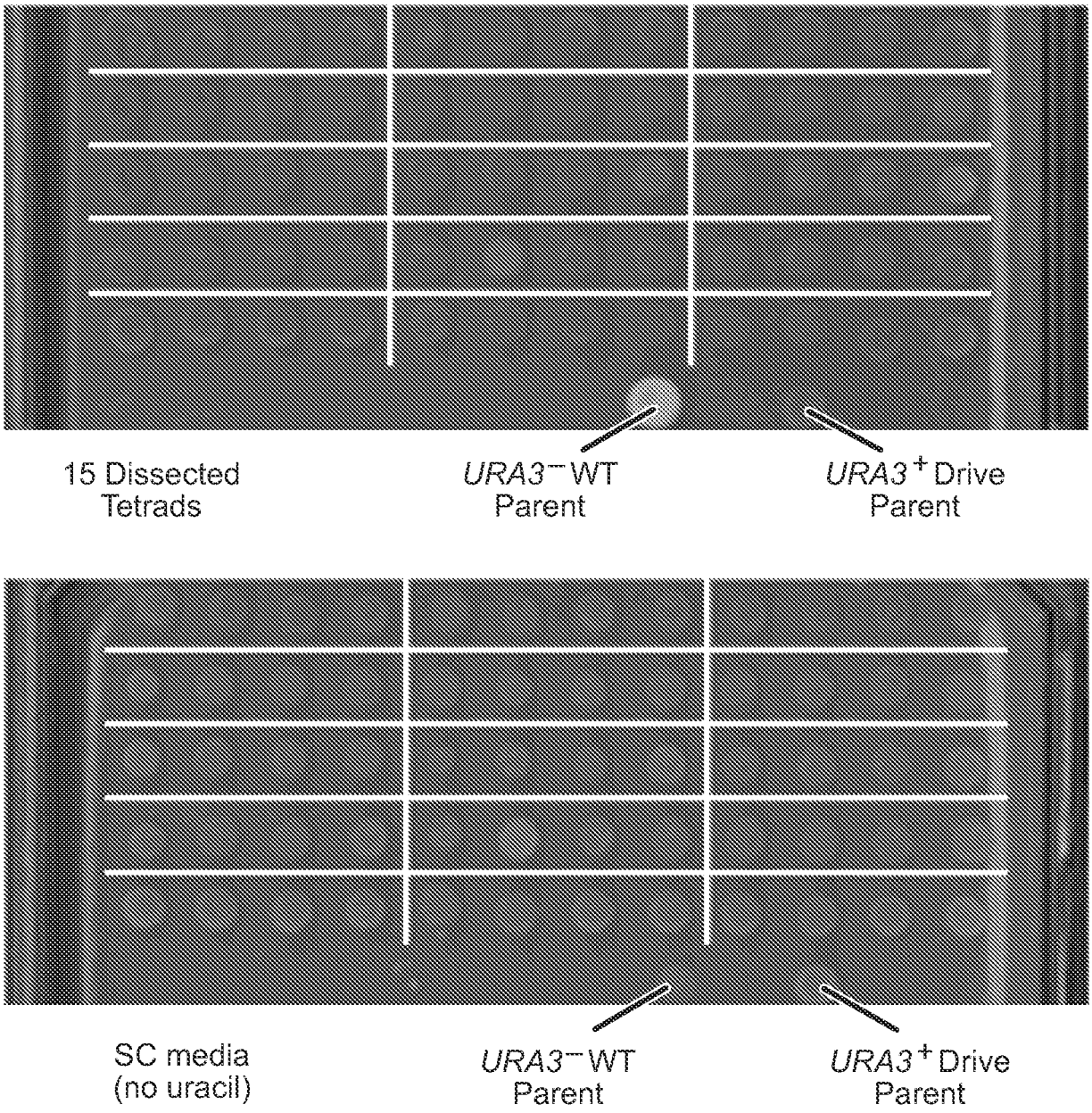

15 Dissected Tetrads
URA3⁻ WT Parent
URA3⁺ Drive Parent
SC media (no uracil)
URA3⁻ WT Parent
URA3⁺ Drive Parent

FIG. 15A

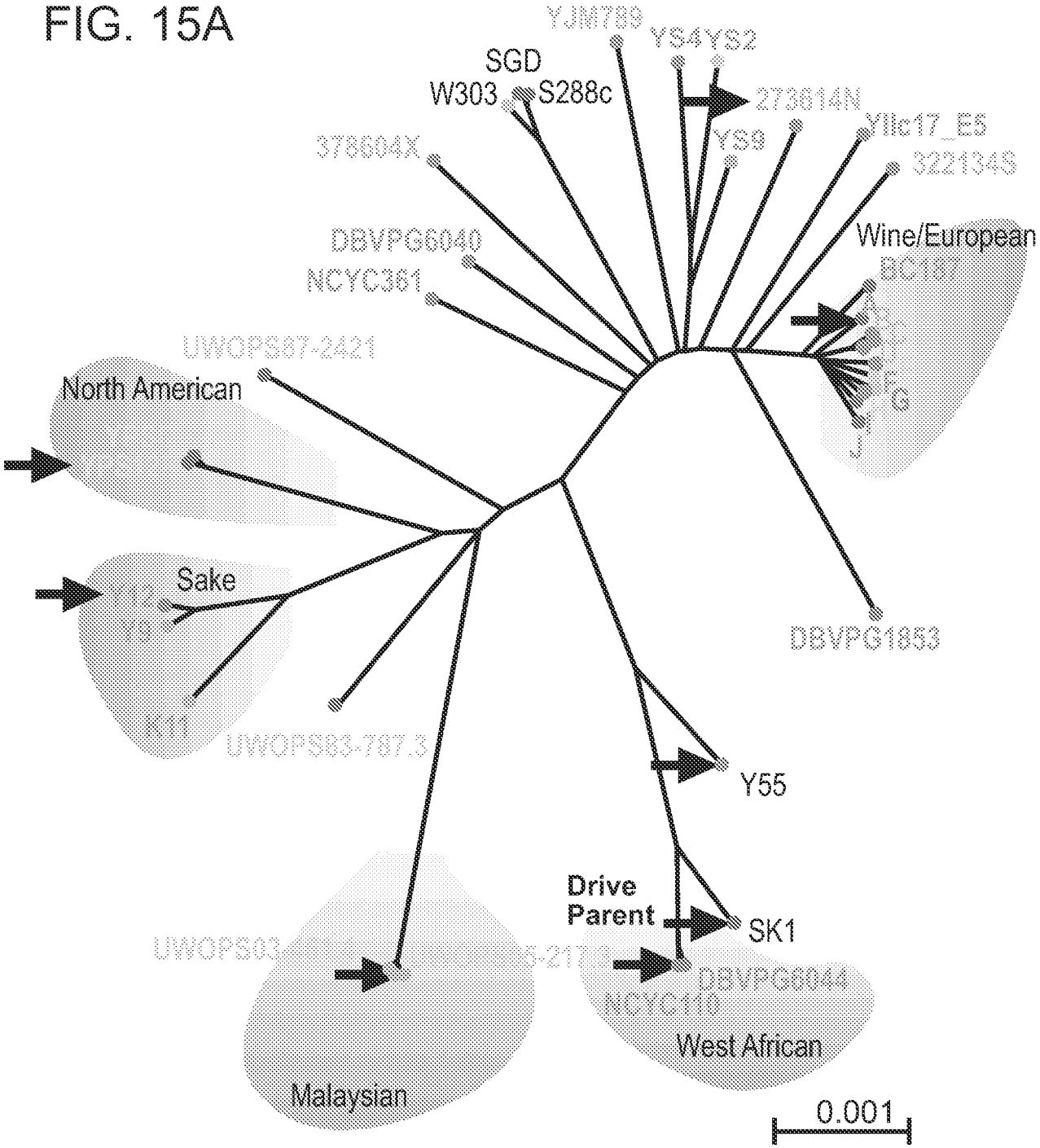
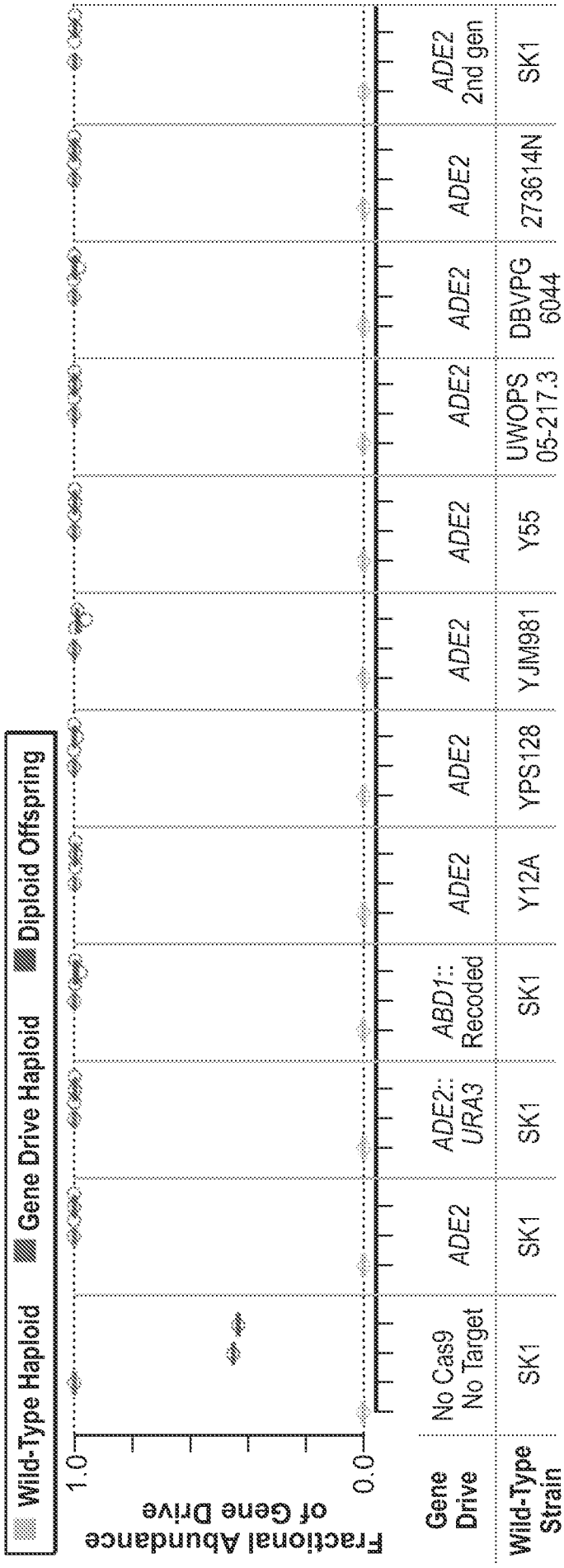

YJM789
YS4YS2
SGD
W303
S288c
273614N
YIIc17_E5
YS9
378604X
322134S
DBVPG6040
NCYC361
Wine/European
UWOPS87-2421
North American
Sake
DBVPG1853
UWOPS83-787.3
Y55
Drive
Parent
SK1
DBVPG6044
NCYC110
West African
Malaysian
0.001
A, RM11_1A
B, YJM981
C, YJM978
D, YJM975
E, DBVPG1373
F, DBVPG1106
G, DBVPG6765
H, DBVPG1788
I, L-1374
J, L-1528
Europe
Americas
Asia
Oceania
Africa
Unknown
Laboratory
Wild
Fermentation
Clinical
Baking
Unknown

FIG. 15B


Wild-Type Haploid
Gene Drive Haploid
Diploid Offspring
Fractional Abundance of Gene Drive
1.0
0.0
Gene Drive
Wild-Type Strain
No Cas9 No Target
SK1
ADE2
SK1
ADE2::URA3
SK1
ABD1:: Recoded
SK1
ADE2
Y12A
ADE2
YPS128
ADE2
YJM981
ADE2
Y55
ADE2
UWOPS 05-217.3
ADE2
DBVPG 6044
ADE2
273614N
ADE2 2nd gen
SK1