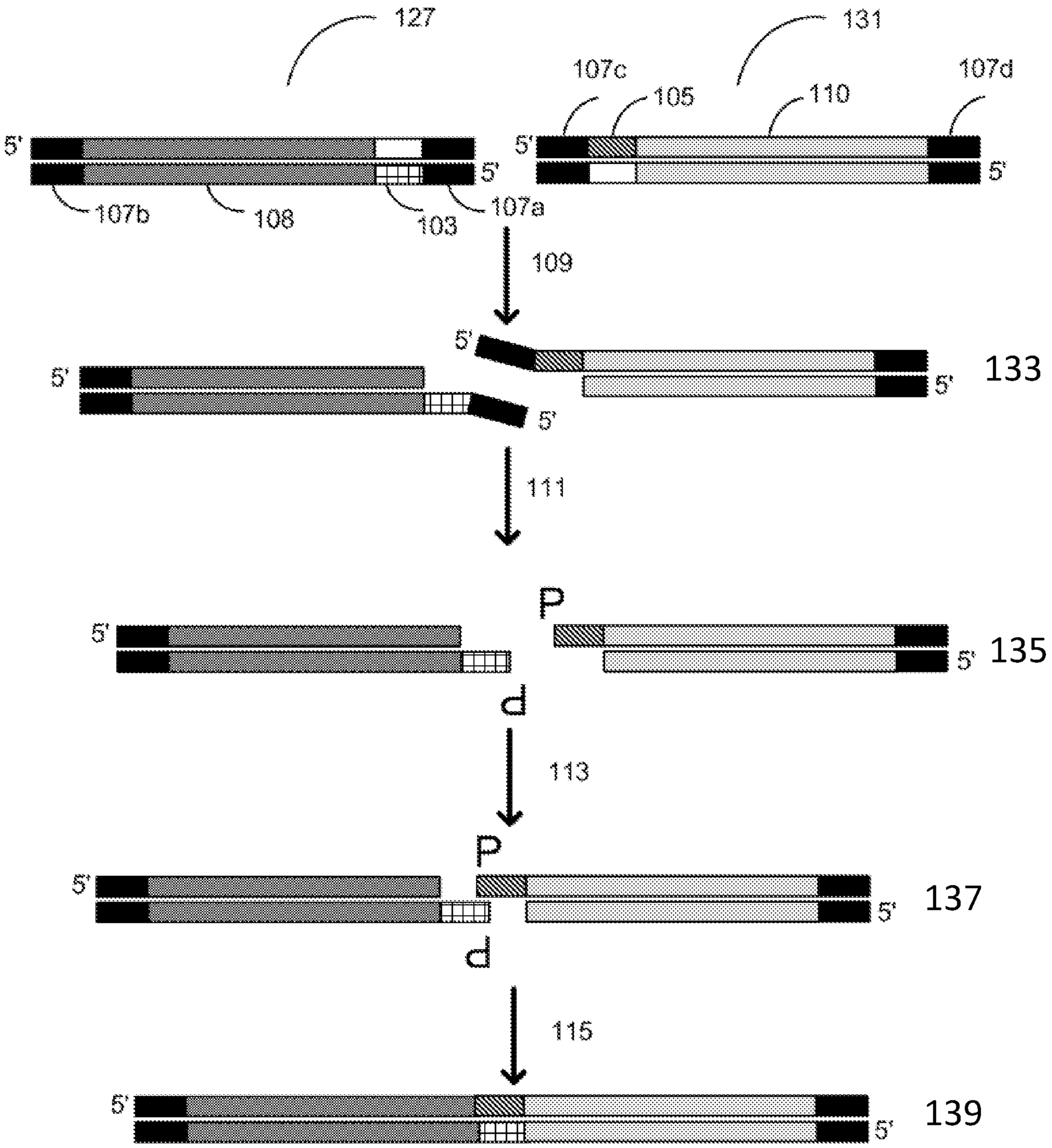

127
5'
107b
108
103
107a
109
131
107c
105
110
107d
5'
5'
133
5'
5'
5'
111
P
5'
135
d
113
P
5'
137
5'
d
115
5'
139
5'

FIG. 1A

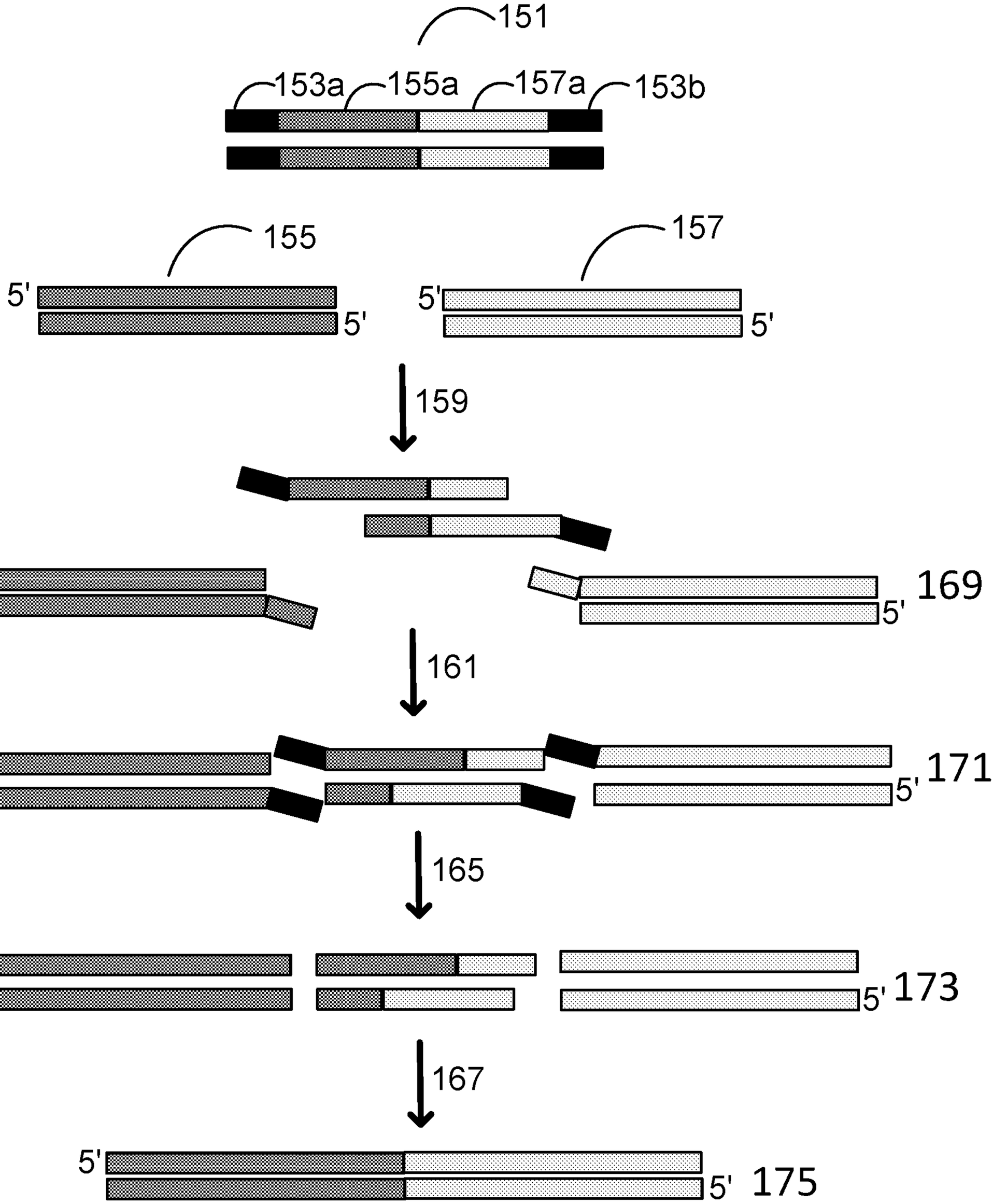

151
153a
155a
157a
153b
155
157
5'
5'
5'
5'
159
169
161
171
165
173
167
175

FIG. 1B

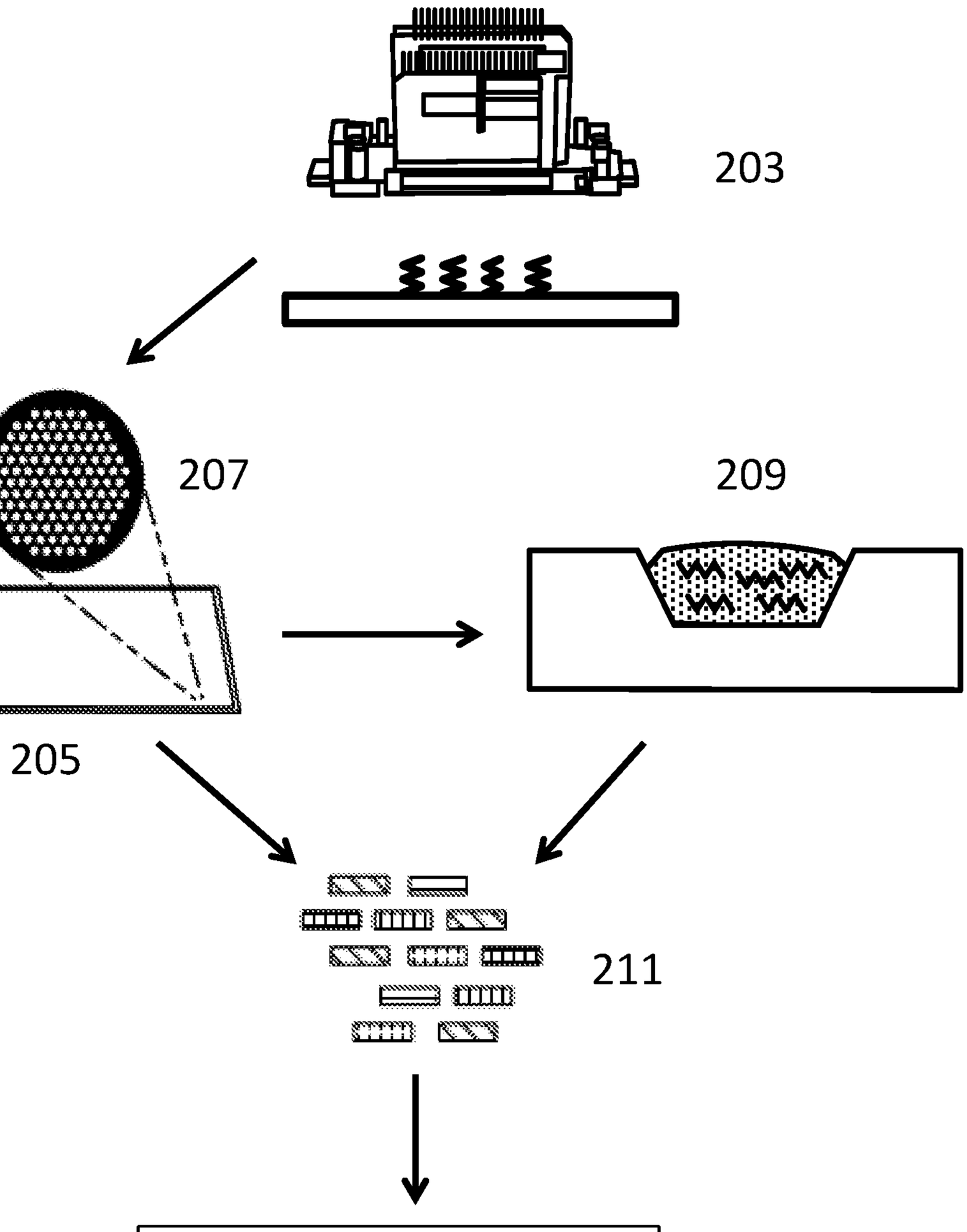

203
207
209
205
211
Flap endonuclease mediated nucleic acid assembly
213

FIG. 2

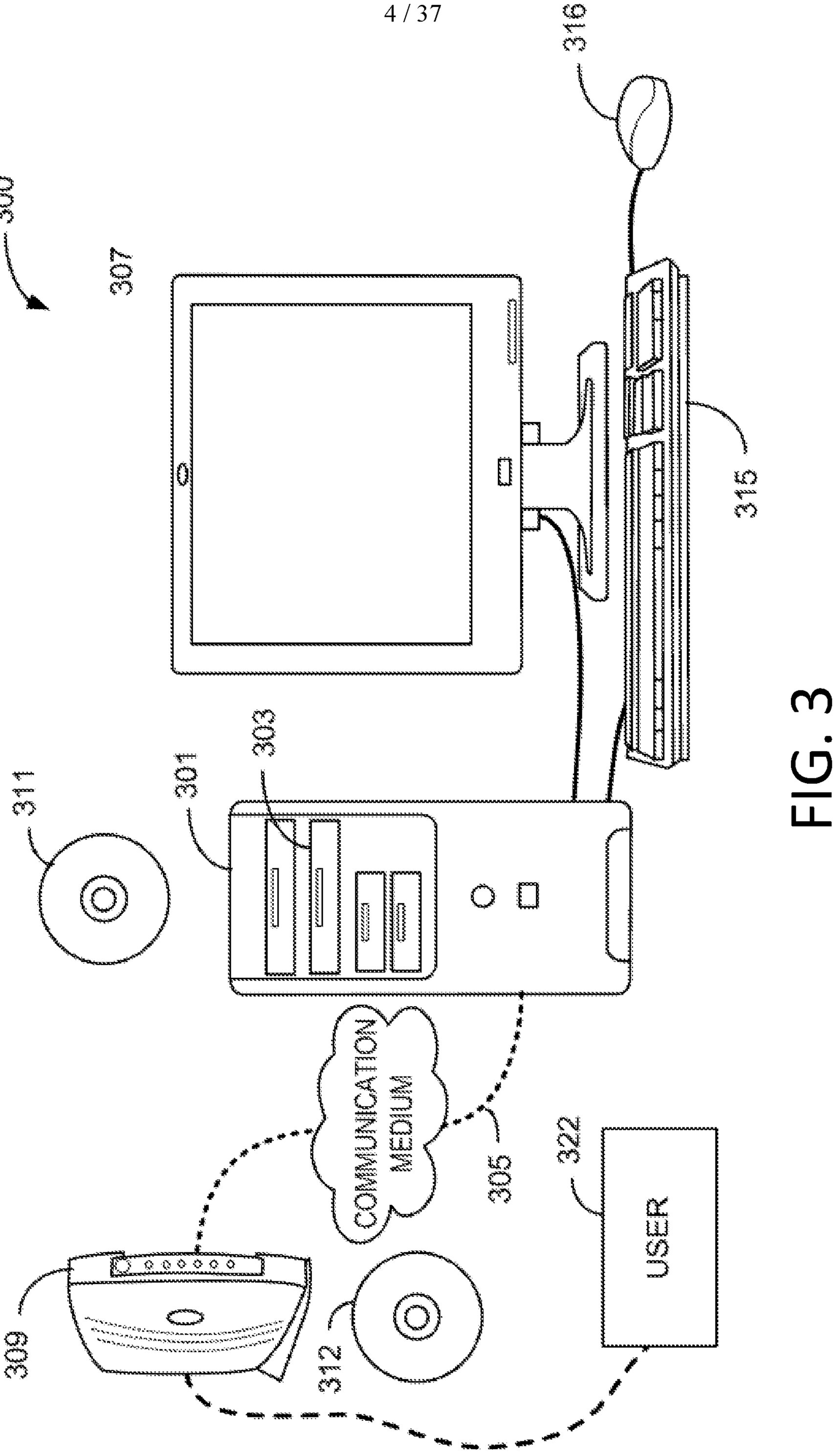

300
307
316
315
311
301
303
305
322
USER
309
312
COMMUNICATION MEDIUM

FIG. 3

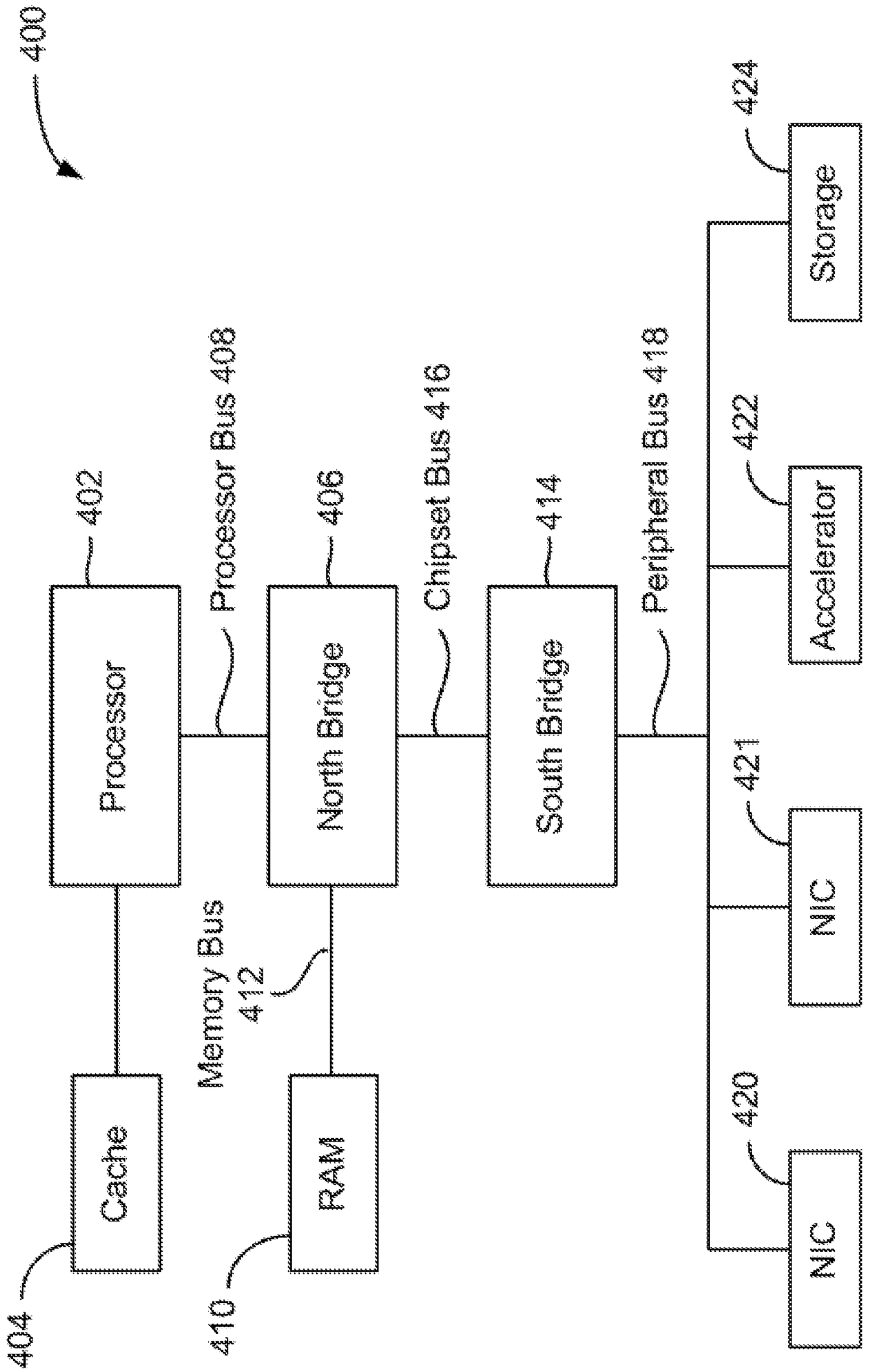

400
402
Processor
Cache
404
Processor Bus 408
406
North Bridge
Memory Bus
412
410
RAM
Chipset Bus 416
414
South Bridge
Peripheral Bus 418
422
Storage
Accelerator
421
NIC
420
NIC

FIG. 4

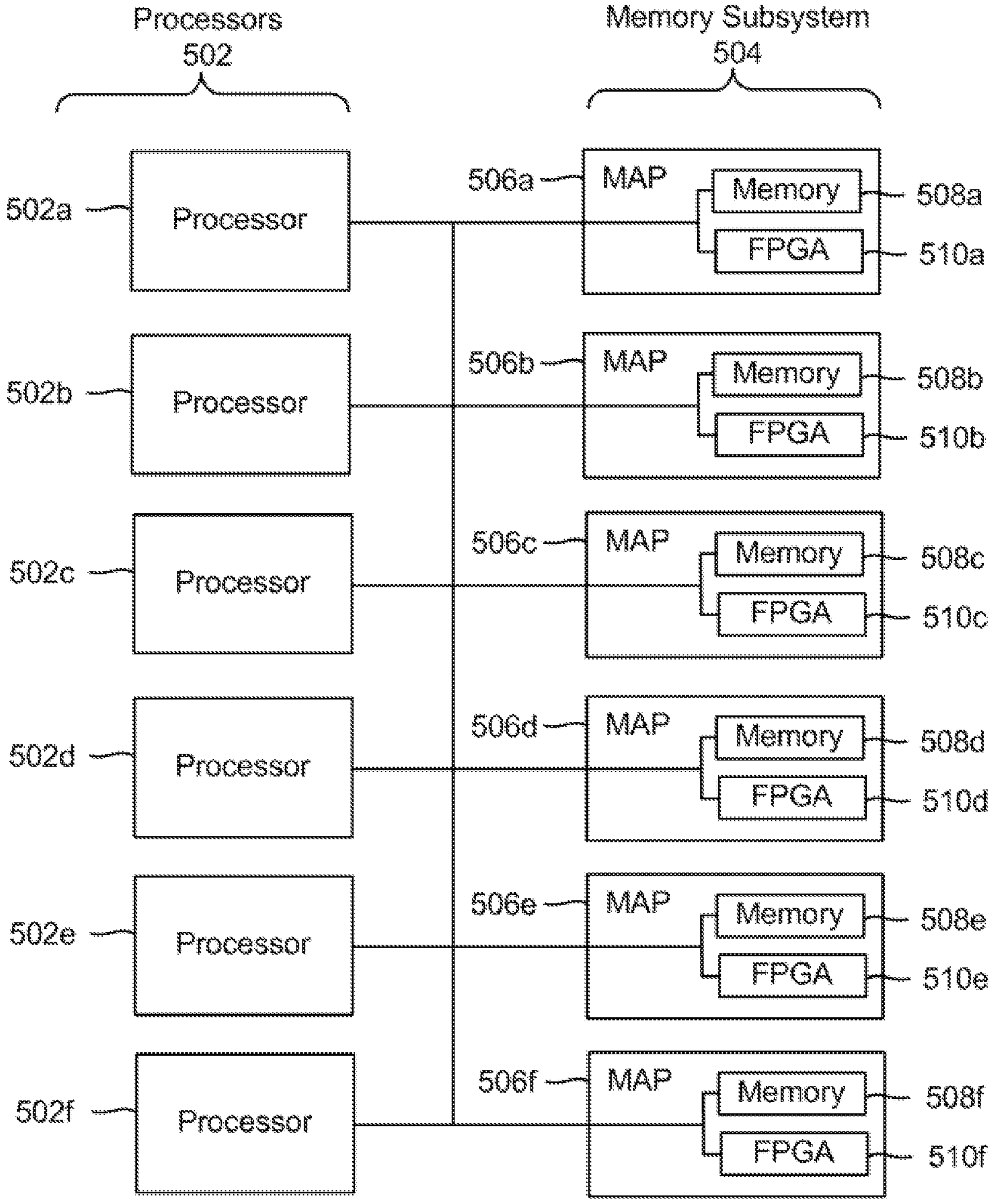

Processors
502
Memory Subsystem
504
502a
Processor
506a
MAP
Memory
508a
FPGA
510a
502b
Processor
506b
MAP
Memory
508b
FPGA
510b
502c
Processor
506c
MAP
Memory
508c
FPGA
510c
502d
Processor
506d
MAP
Memory
508d
FPGA
510d
502e
Processor
506e
MAP
Memory
508e
FPGA
510e
502f
Processor
506f
MAP
Memory
508f
FPGA
510f

FIG. 5

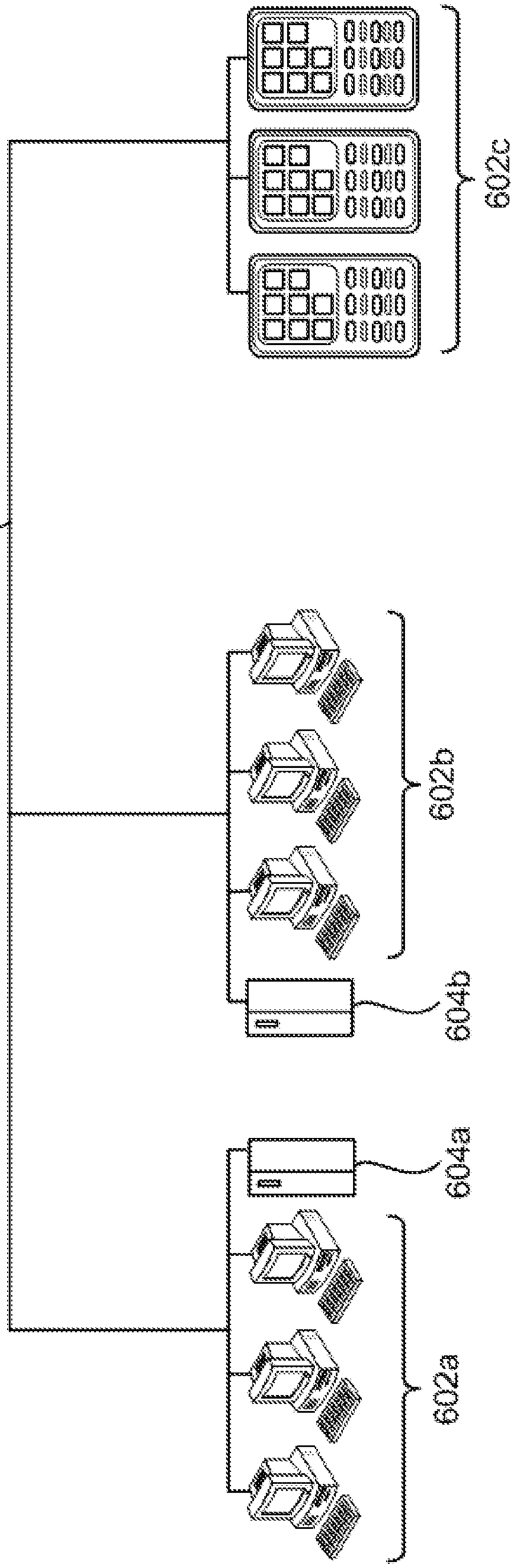


FIG. 6

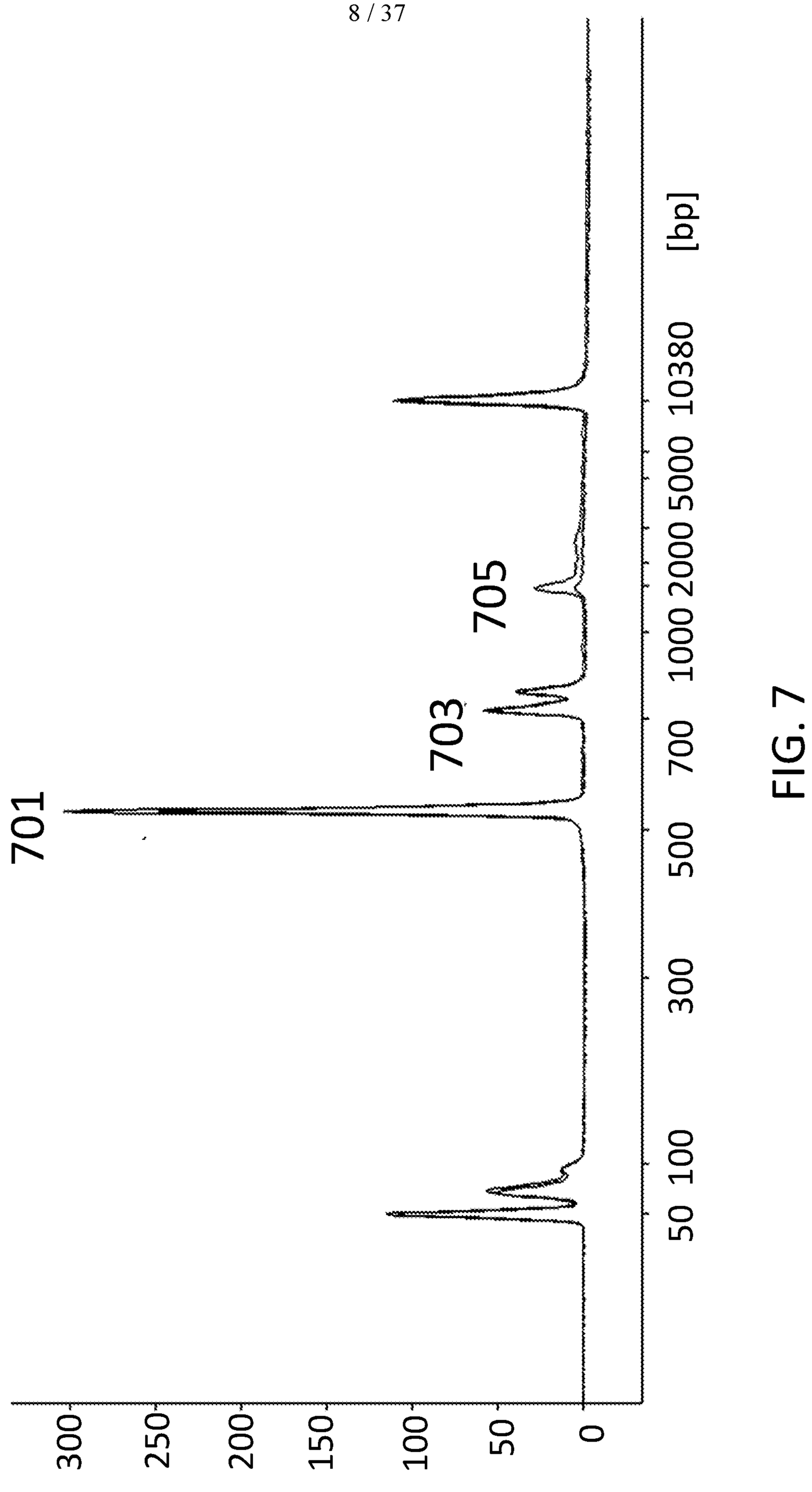

[FU]
300
250
200
150
100
50
0
701
703
705
50
100
300
500
700
1000
2000
5000
10380
[bp]

FIG. 7

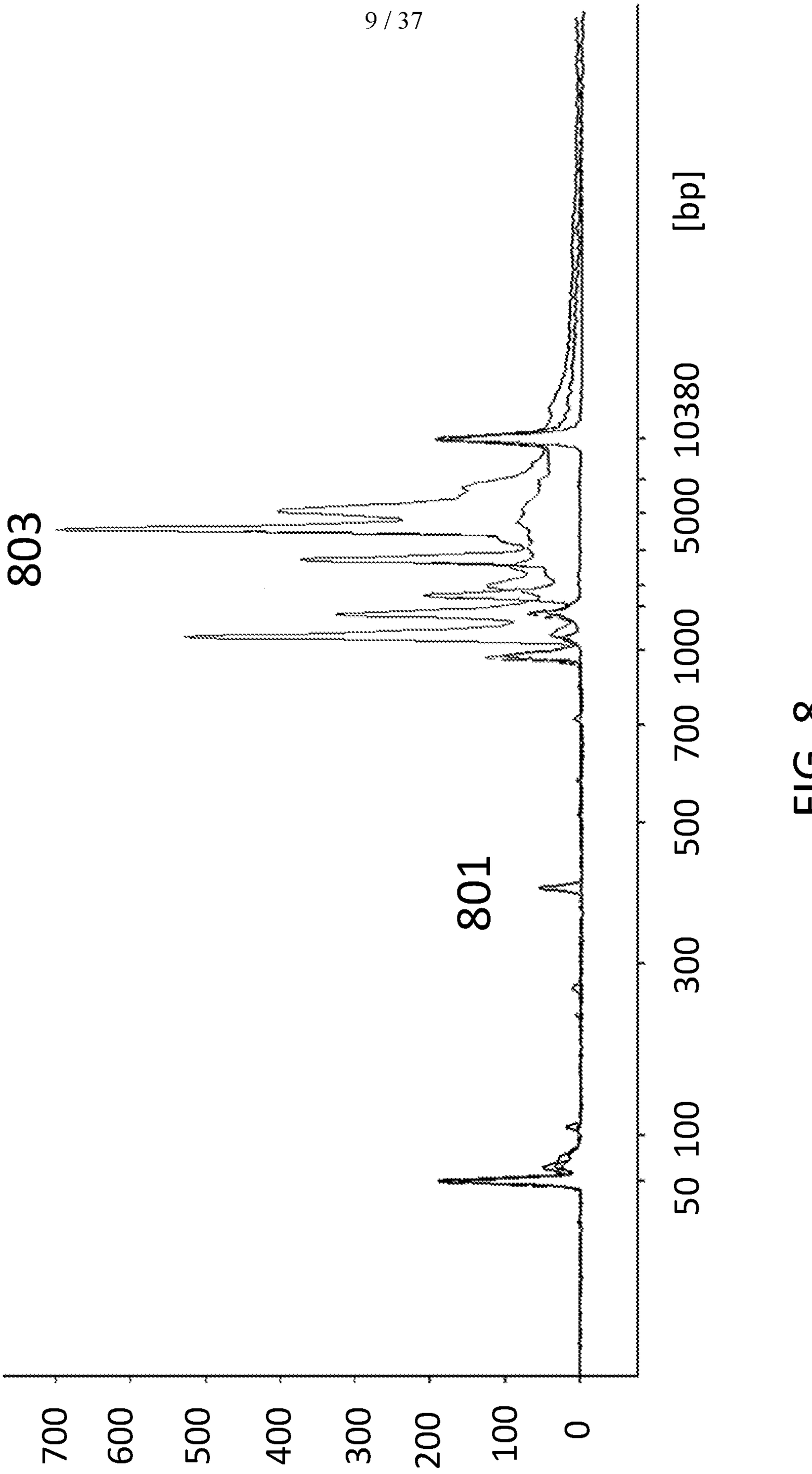

[pa]
803
801
[FU]
700
600
500
400
300
200
100
0
50
100
300
500
700
1000
5000
10380

FIG. 8

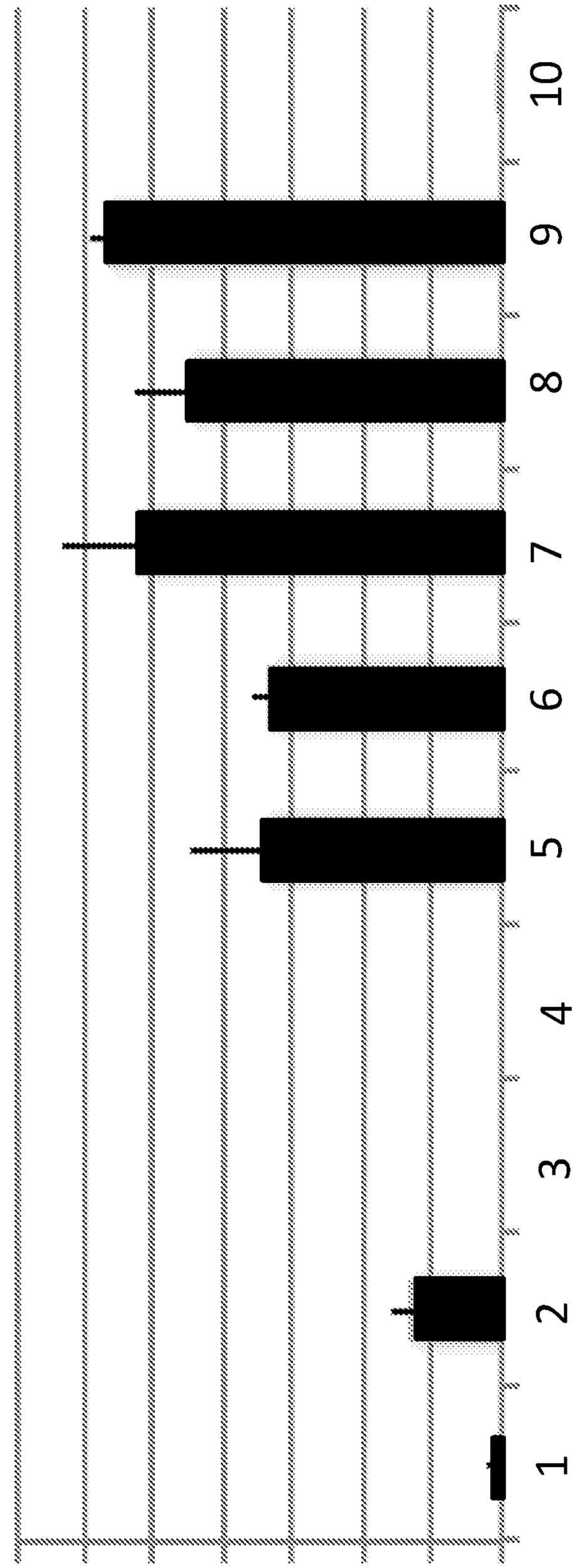

10
9
8
7
6
5
4
3
2
1
0
10000
20000
30000
40000
50000
60000
70000
CFU

FIG. 9

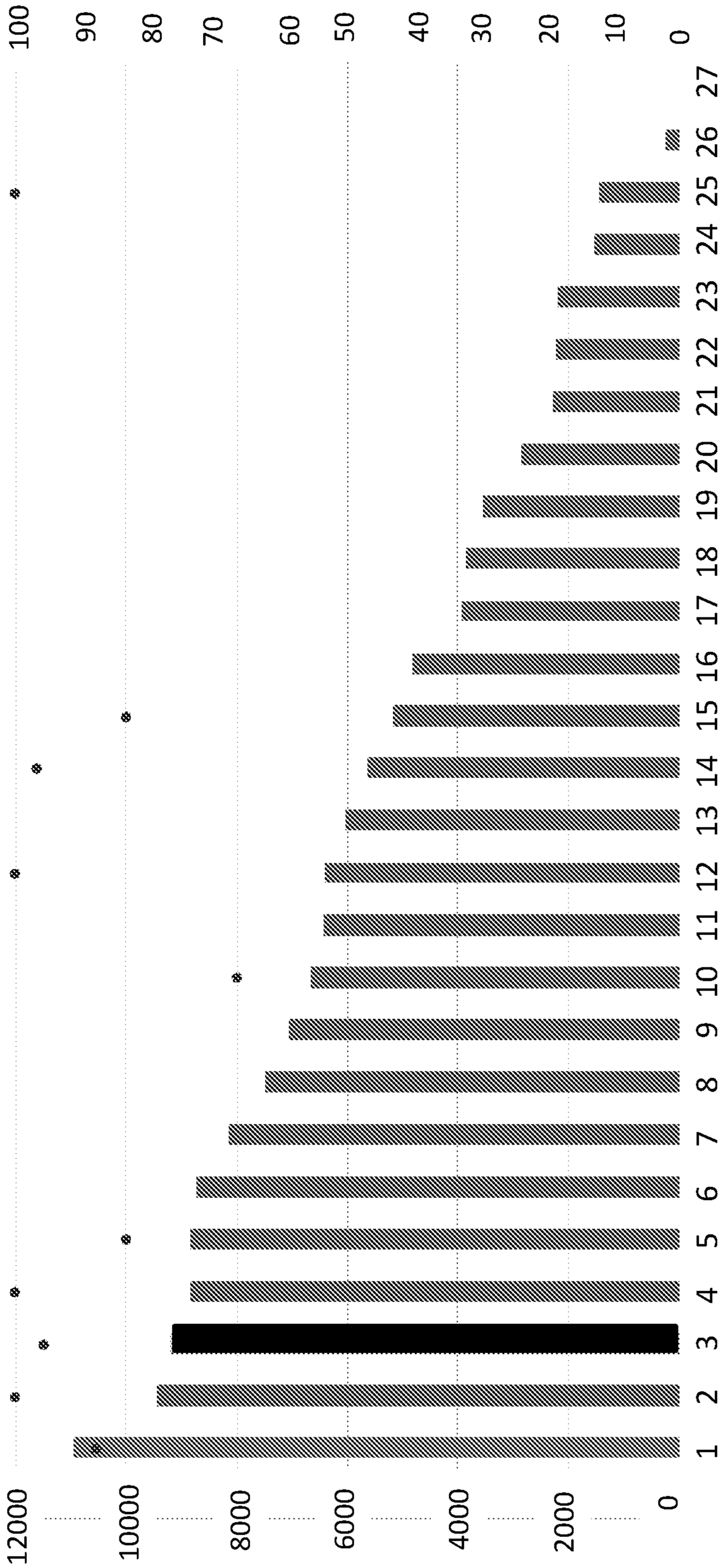

% Correct (measured by cPCR)
100
90
80
70
60
50
40
30
20
10
0
26
27
25
24
23
22
21
20
19
18
17
16
15
14
13
12
11
10
9
8
7
6
5
4
3
2
1
12000
10000
8000
6000
4000
2000
0
Colony Forming Unit (CFU)

FIG. 10

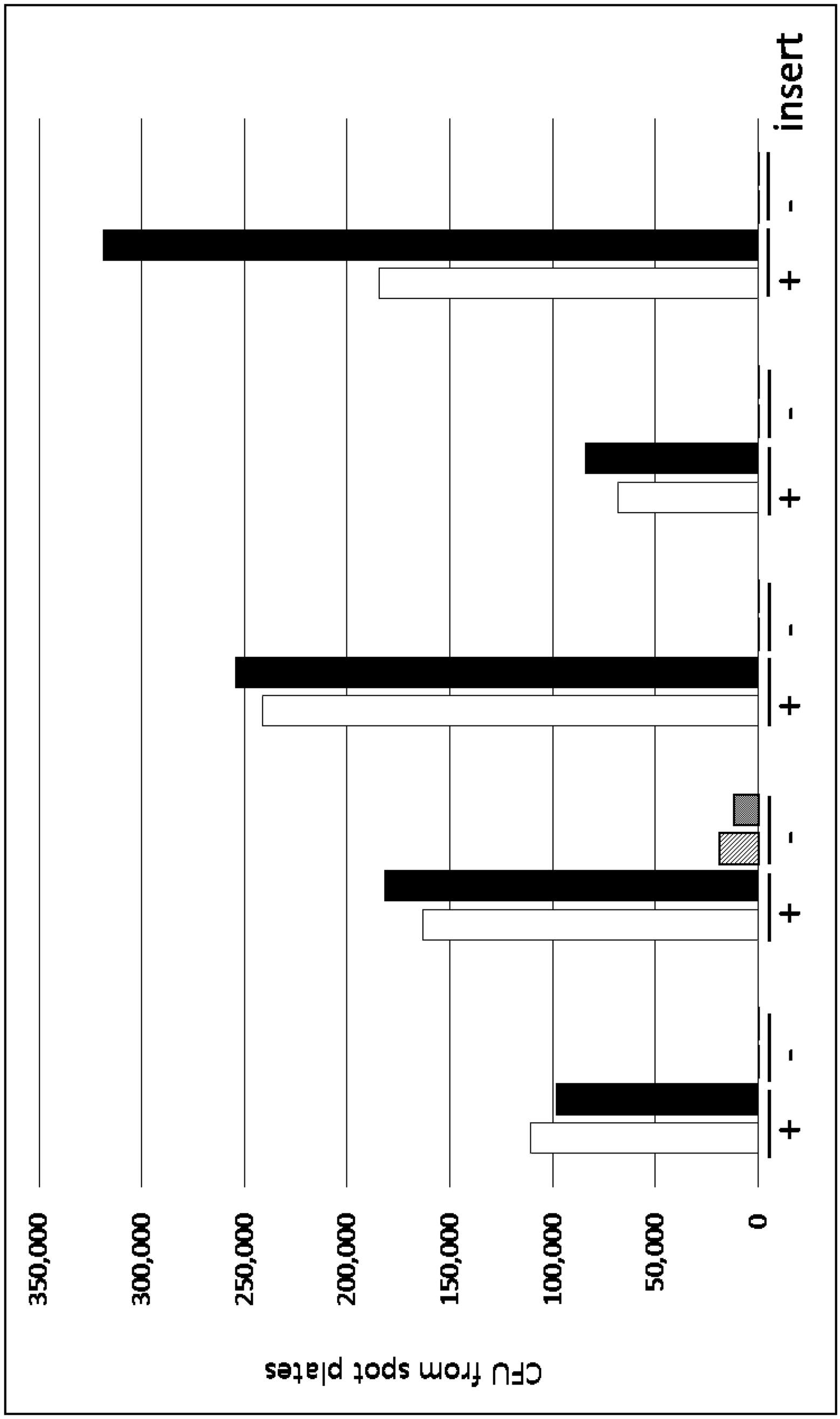

insert
-
+
-
+
-
+
-
+
-
+
CFU from spot plates
350,000
300,000
250,000
200,000
150,000
100,000
50,000
0

FIG. 11

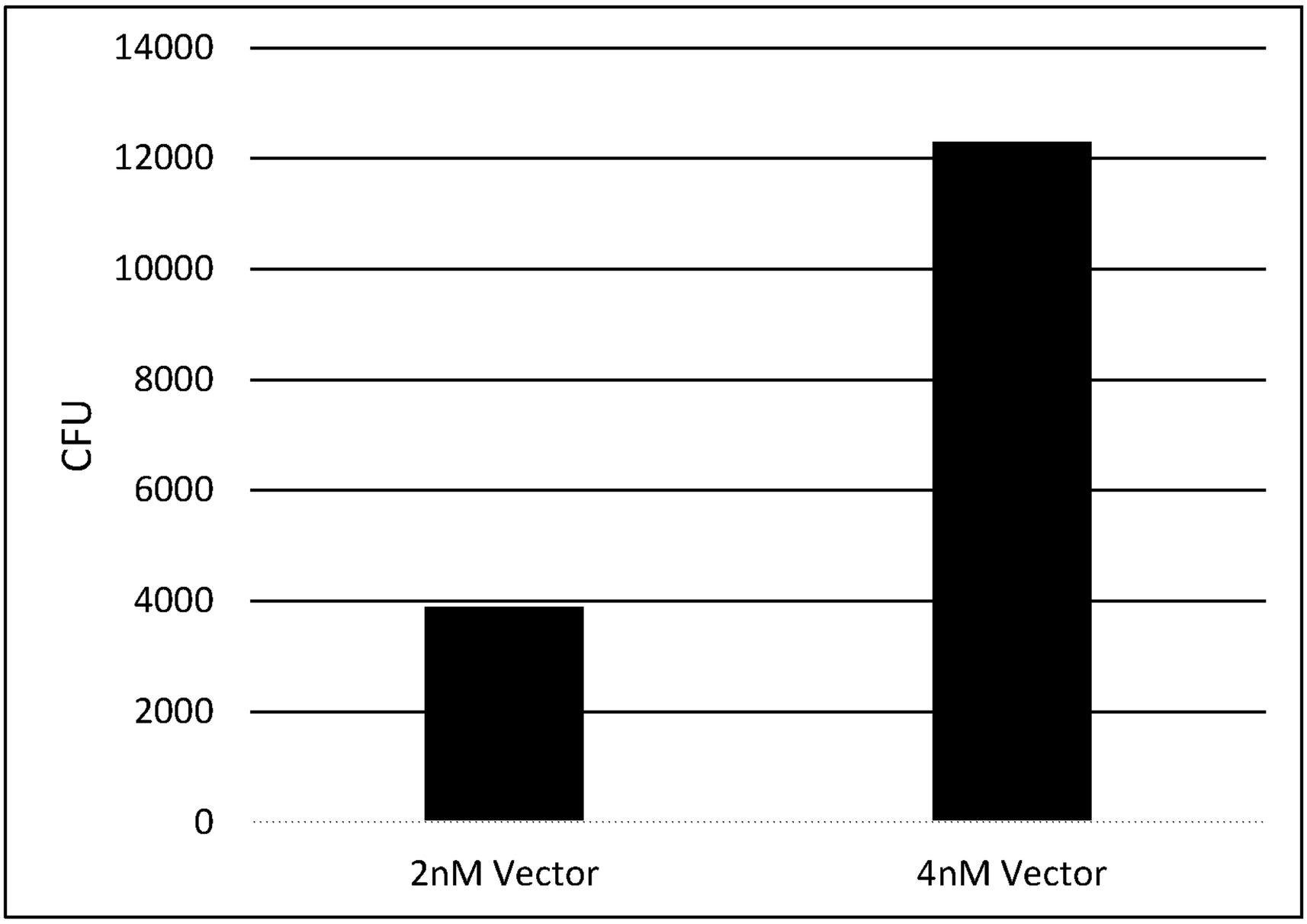

14000
12000
10000
8000
6000
4000
2000
0
CFU
2nM Vector
4nM Vector

FIG. 12

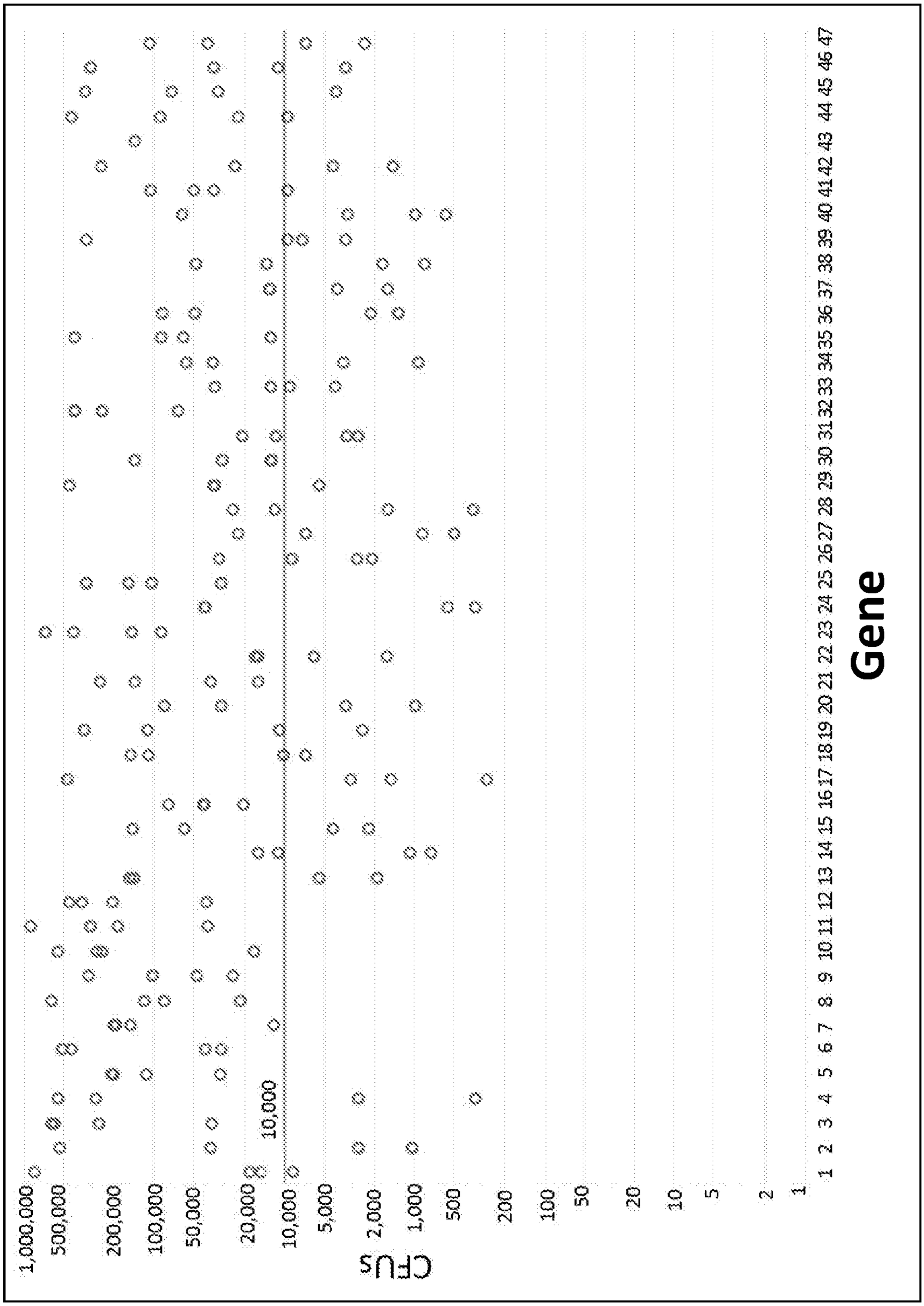

CFUs
1,000,000
500,000
200,000
100,000
50,000
20,000
10,000
5,000
2,000
1,000
500
200
100
50
20
10
5
2
1
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47
Gene

FIG. 13

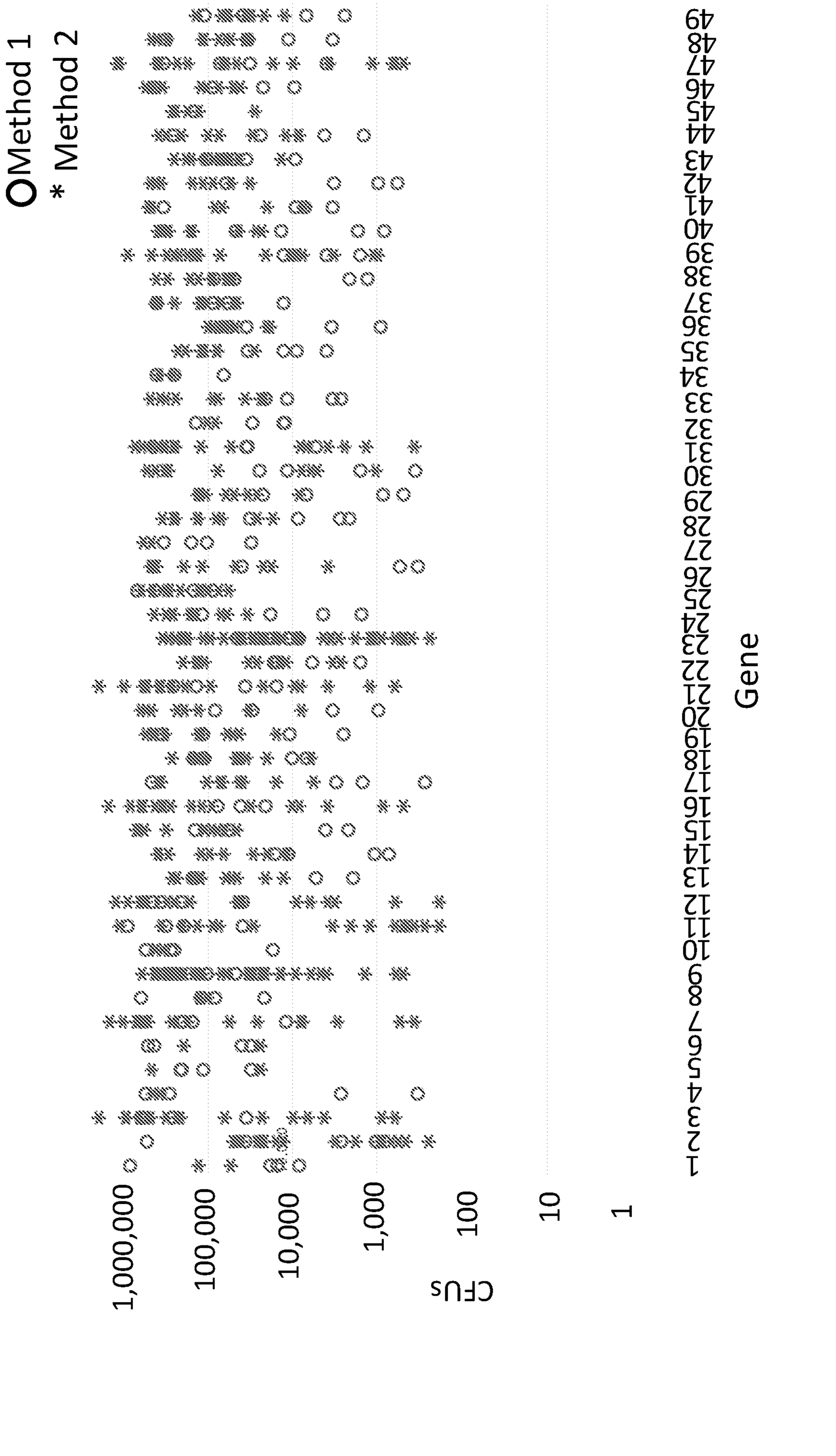

Method 1
Method 2
CFUs
1,000,000
100,000
10,000
1,000
100
10
1
Gene
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 42 43 44 45 46 47 48 49

FIG. 14A

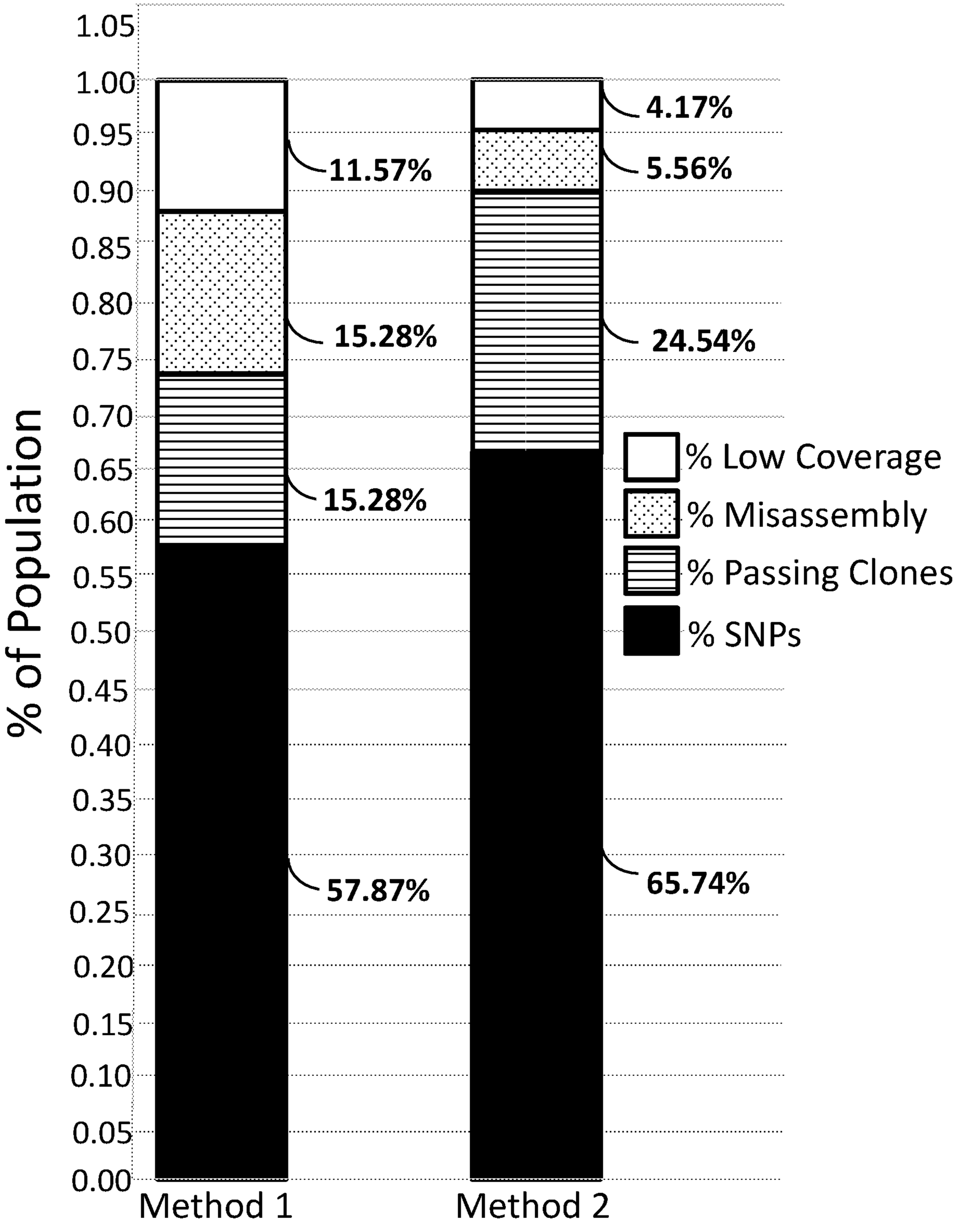

1.05
1.00
0.95
0.90
0.85
0.80
0.75
0.70
0.65
0.60
0.55
0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
% of Population
11.57%
15.28%
15.28%
57.87%
4.17%
5.56%
24.54%
65.74%
% Low Coverage
% Misassembly
% Passing Clones
% SNPs
Method 1
Method 2

FIG. 14B

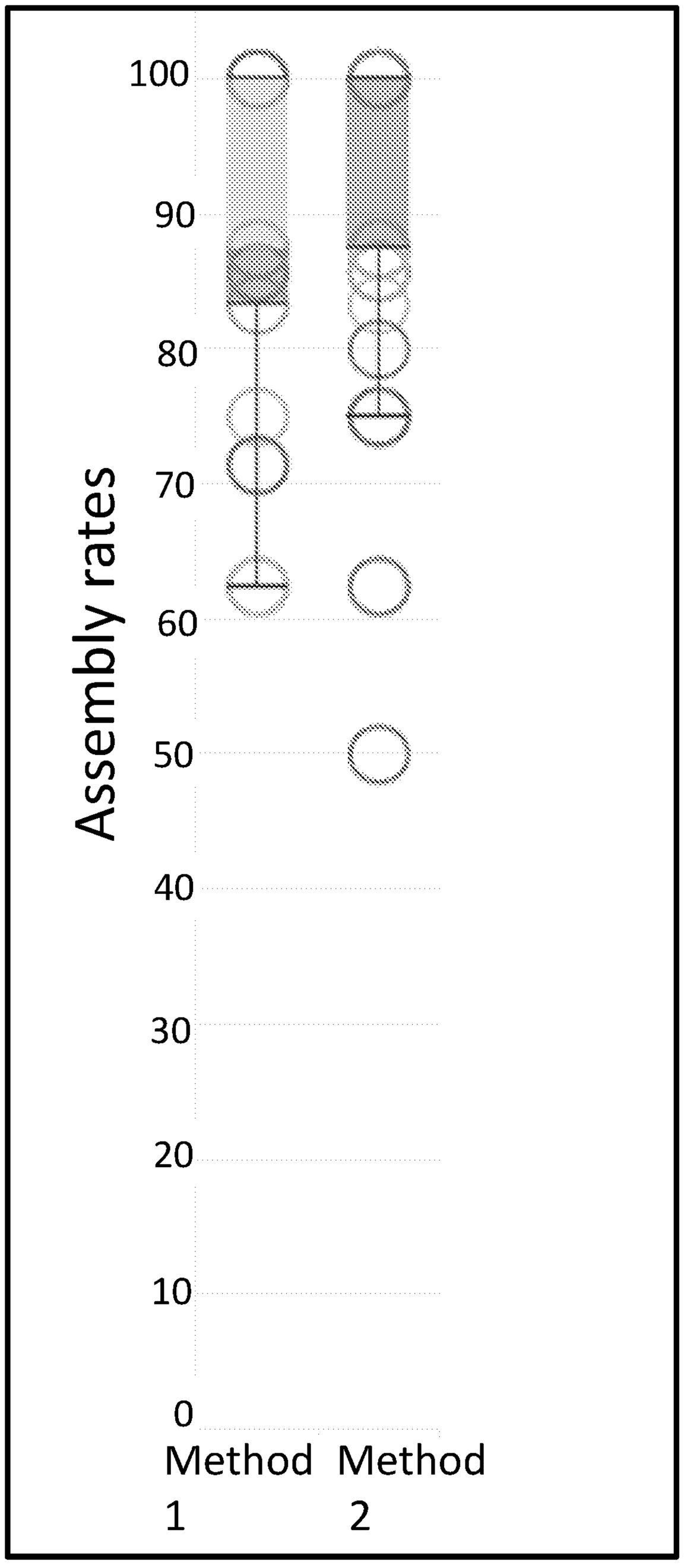


FIG. 14C

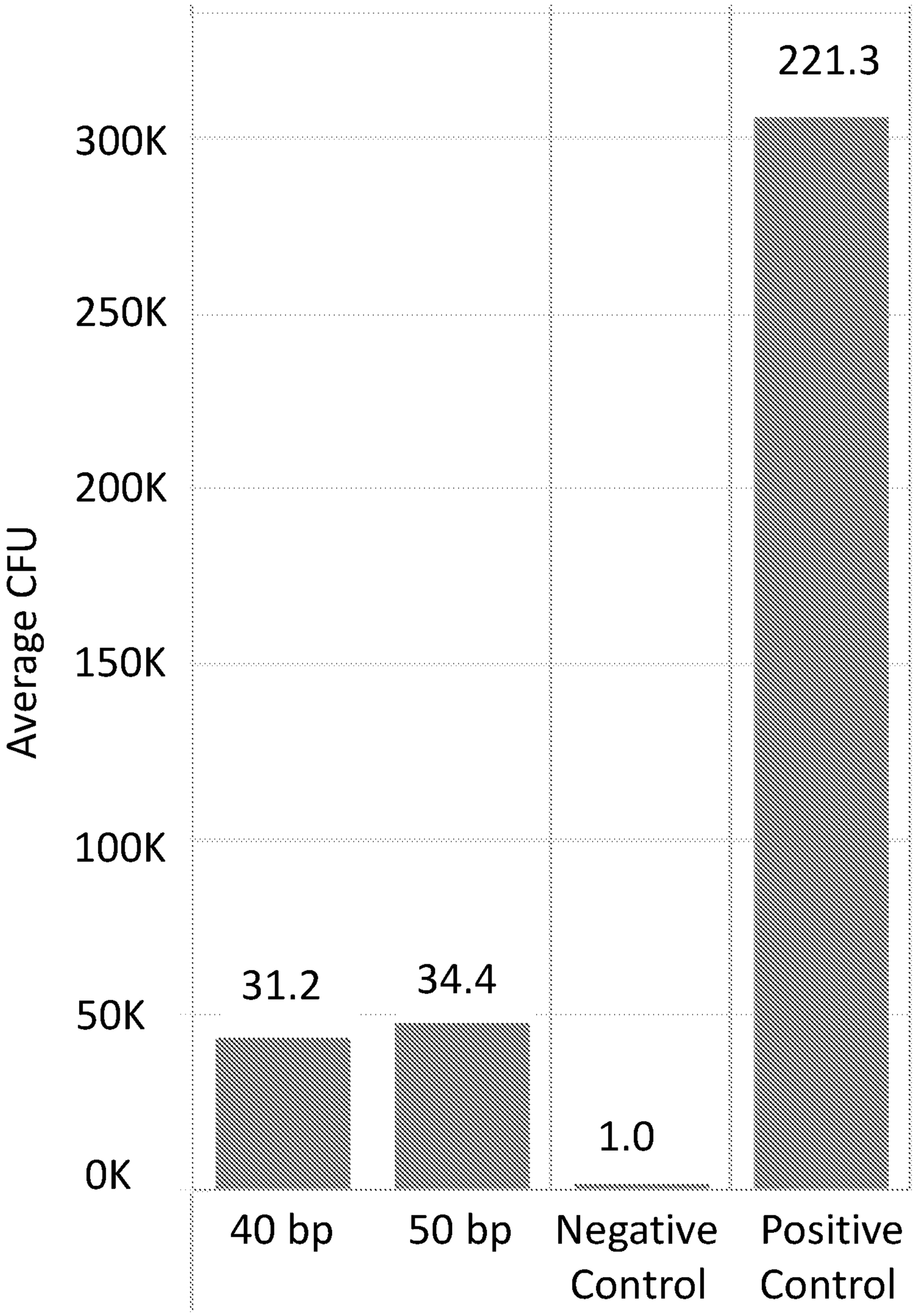

Average CFU
300K
250K
200K
150K
100K
50K
0K
31.2
34.4
1.0
221.3
40 bp
50 bp
Negative Control
Positive Control

FIG. 15A

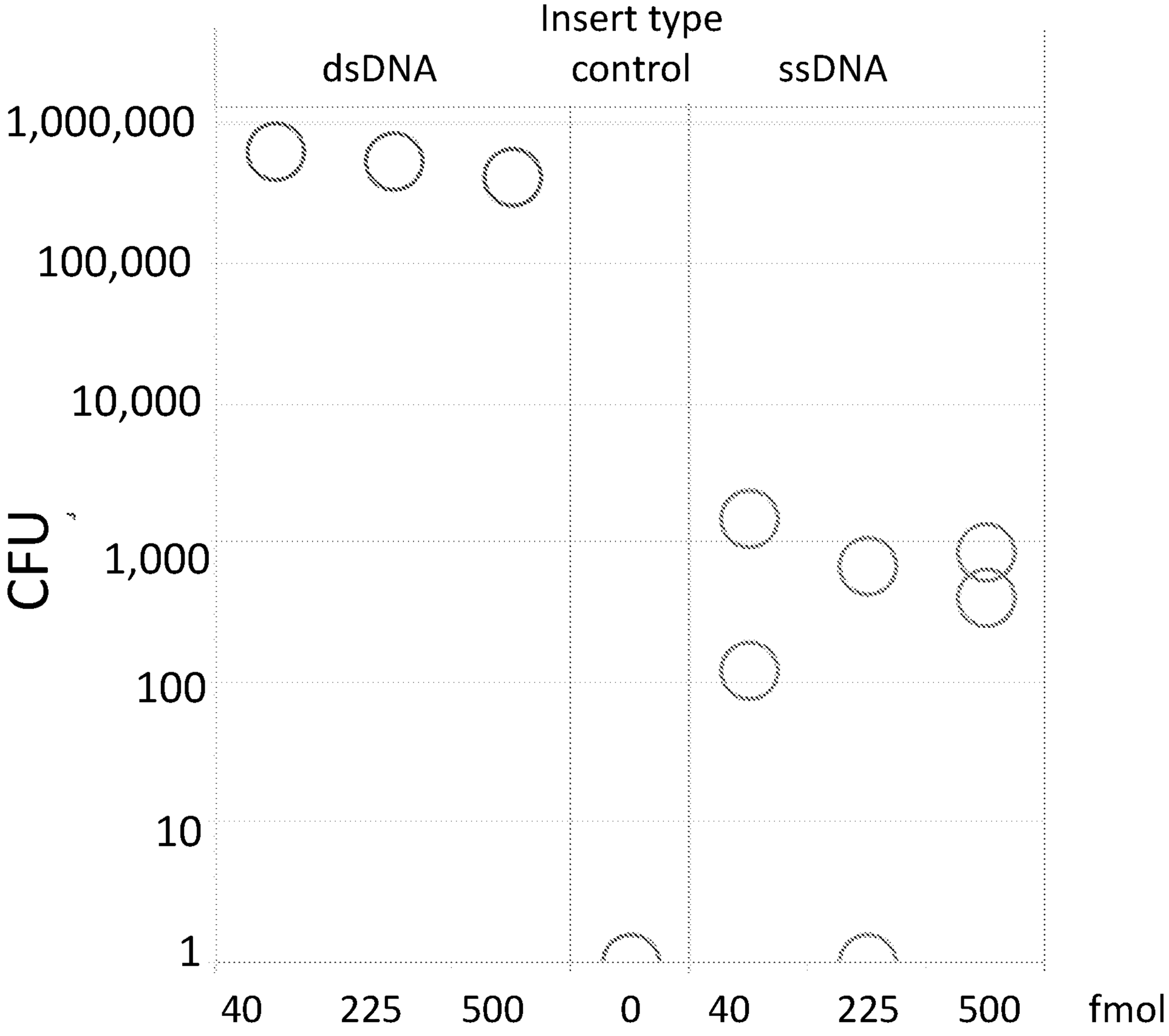

Insert type
dsDNA
control
ssDNA
1,000,000
100,000
10,000
1,000
100
10
1
CFU
40
225
500
0
40
225
500
fmol

FIG. 15B

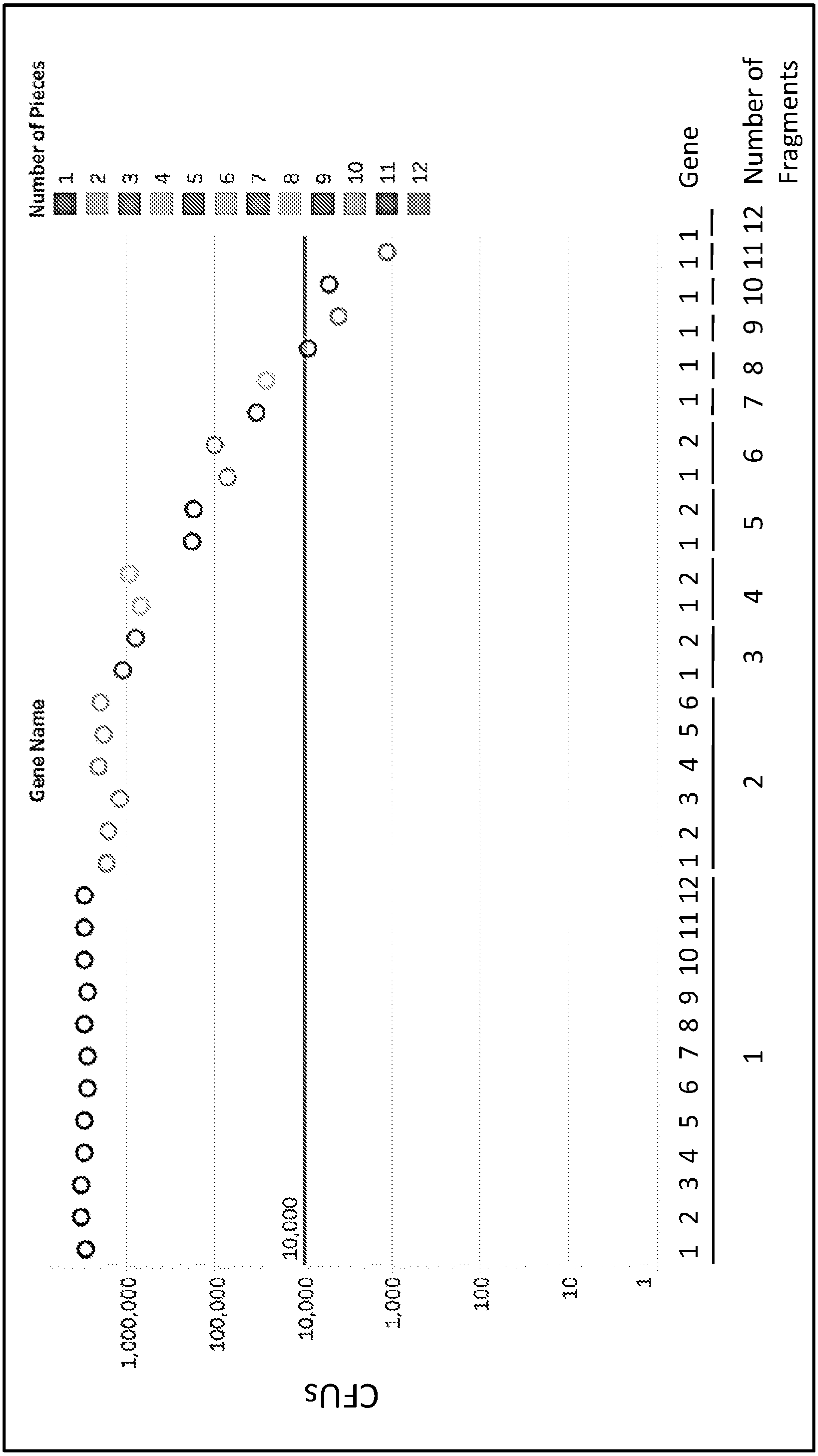

Number of Pieces
1 2 3 4 5 6 7 8 9 10 11 12
Gene Name
CFUs
1,000,000
100,000
10,000
1,000
100
10
1
10,000
1 2 3 4 5 6 7 8 9 10 11 12 1 2 3 4 5 6 1 2 1 2 1 2 1 2 1 1 1 1 1 1 Gene
1 2 3 4 5 6 7 8 9 10 11 12 Number of Fragments

FIG. 16A

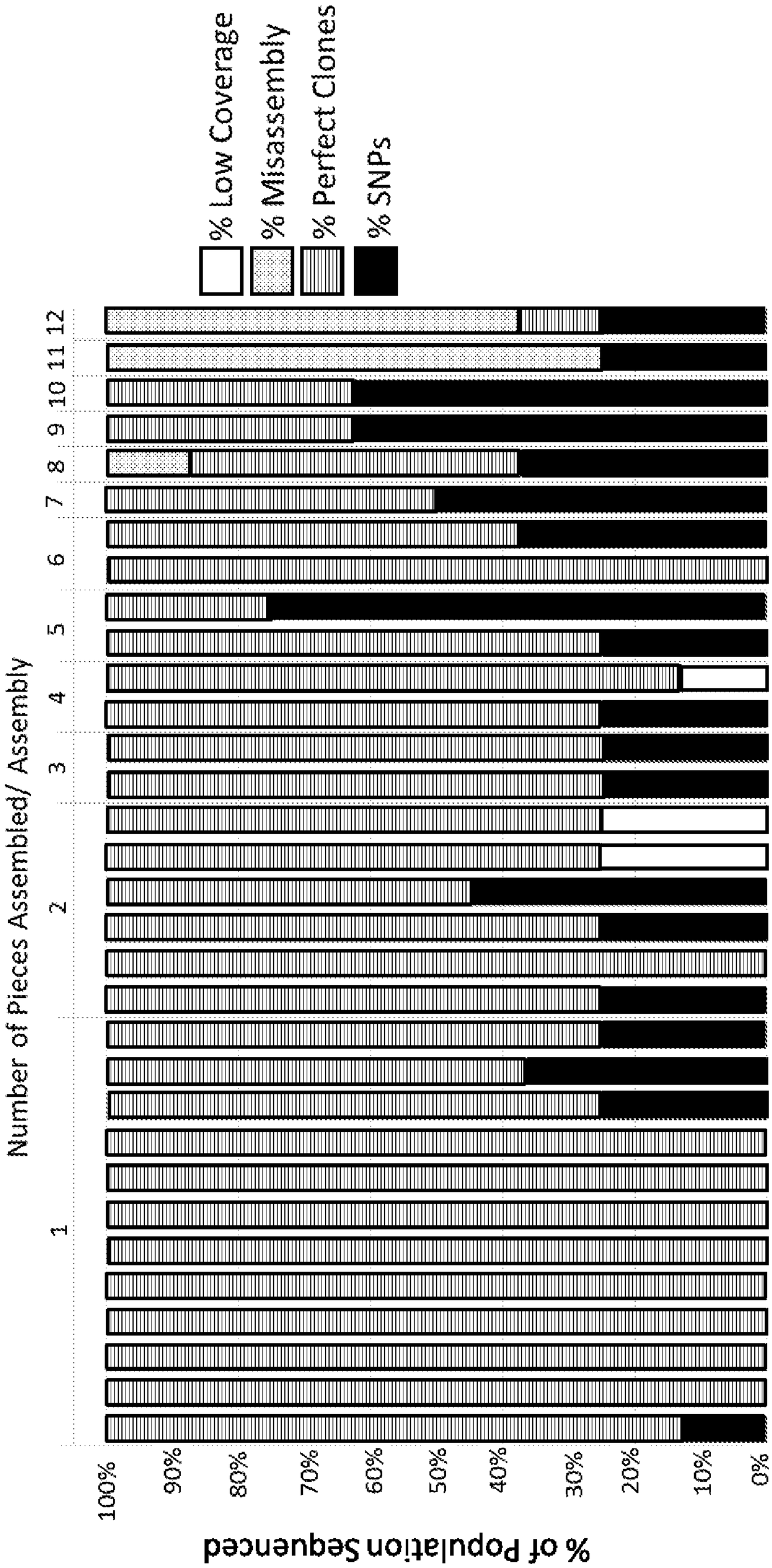

Low Coverage
Misassembly
Perfect Clones
SNPs
% % % %
12
11
10
9
8
7
6
5
4
3
2
1
Number of Pieces Assembled/ Assembly
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
% of Population Sequenced

FIG. 16B

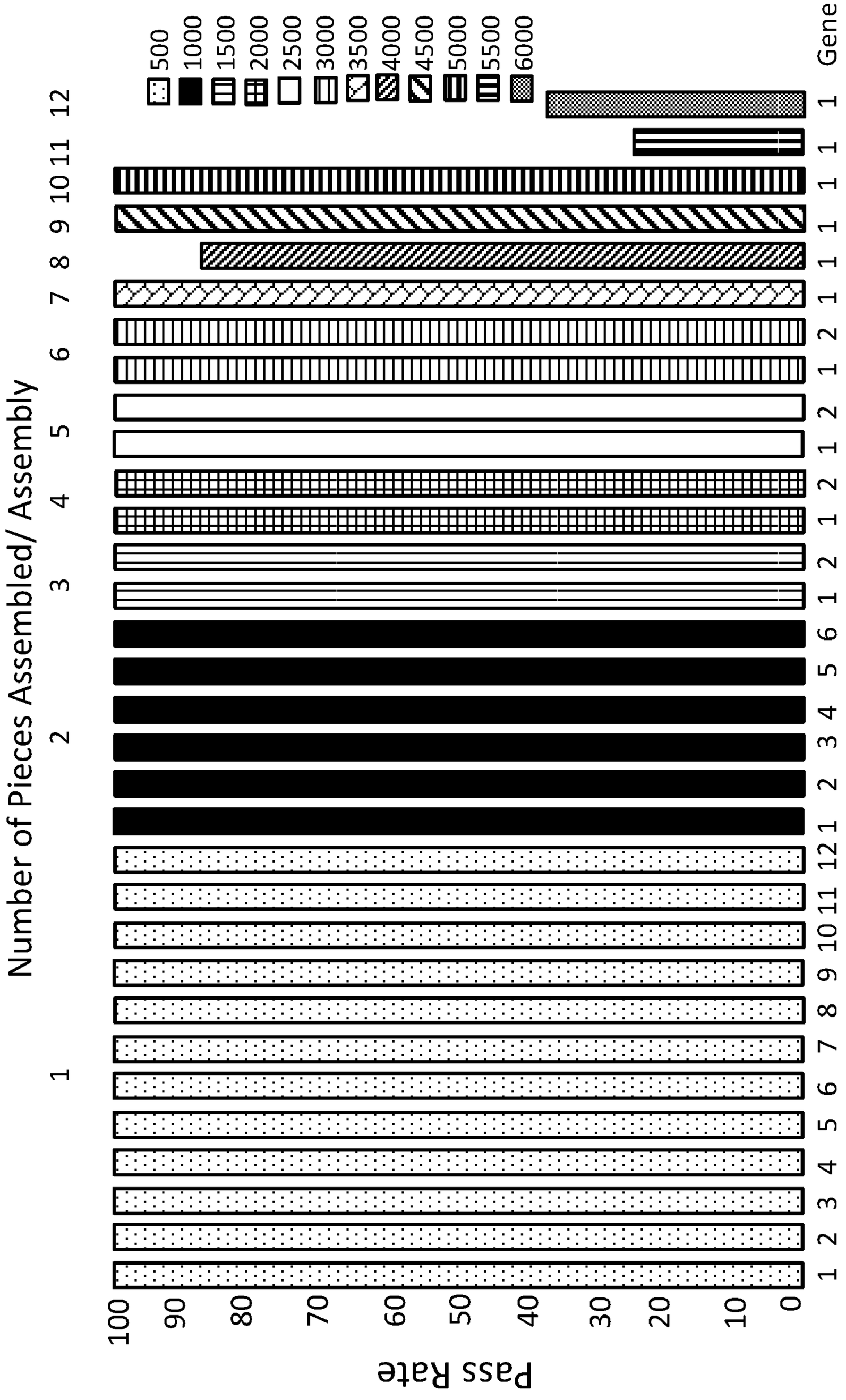

Pass Rate
100
90
80
70
60
50
40
30
20
10
0
Number of Pieces Assembled/ Assembly
1
2
3
4
5
6
7
8
9
10
11
12
Gene
500
1000
1500
2000
2500
3000
3500
4000
4500
5000
5500
6000

FIG. 16C

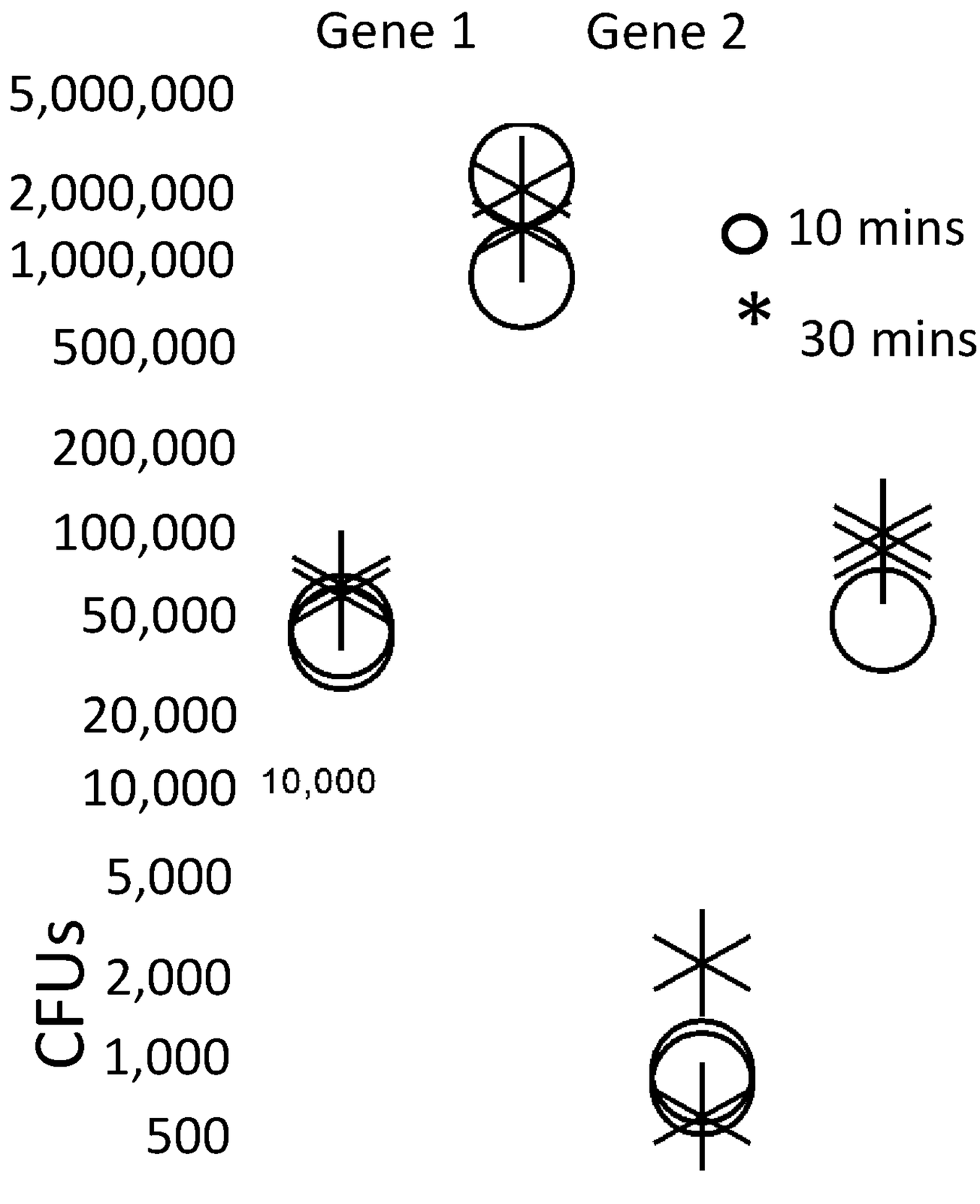

Gene 1
Gene 2
5,000,000
2,000,000
1,000,000
500,000
200,000
100,000
50,000
20,000
10,000
10,000
5,000
2,000
1,000
500
200
100
50
20
10
5
2
1
CFUs
10 mins
30 mins
Method 2
Method 3
Method 2
Method 3

FIG. 17

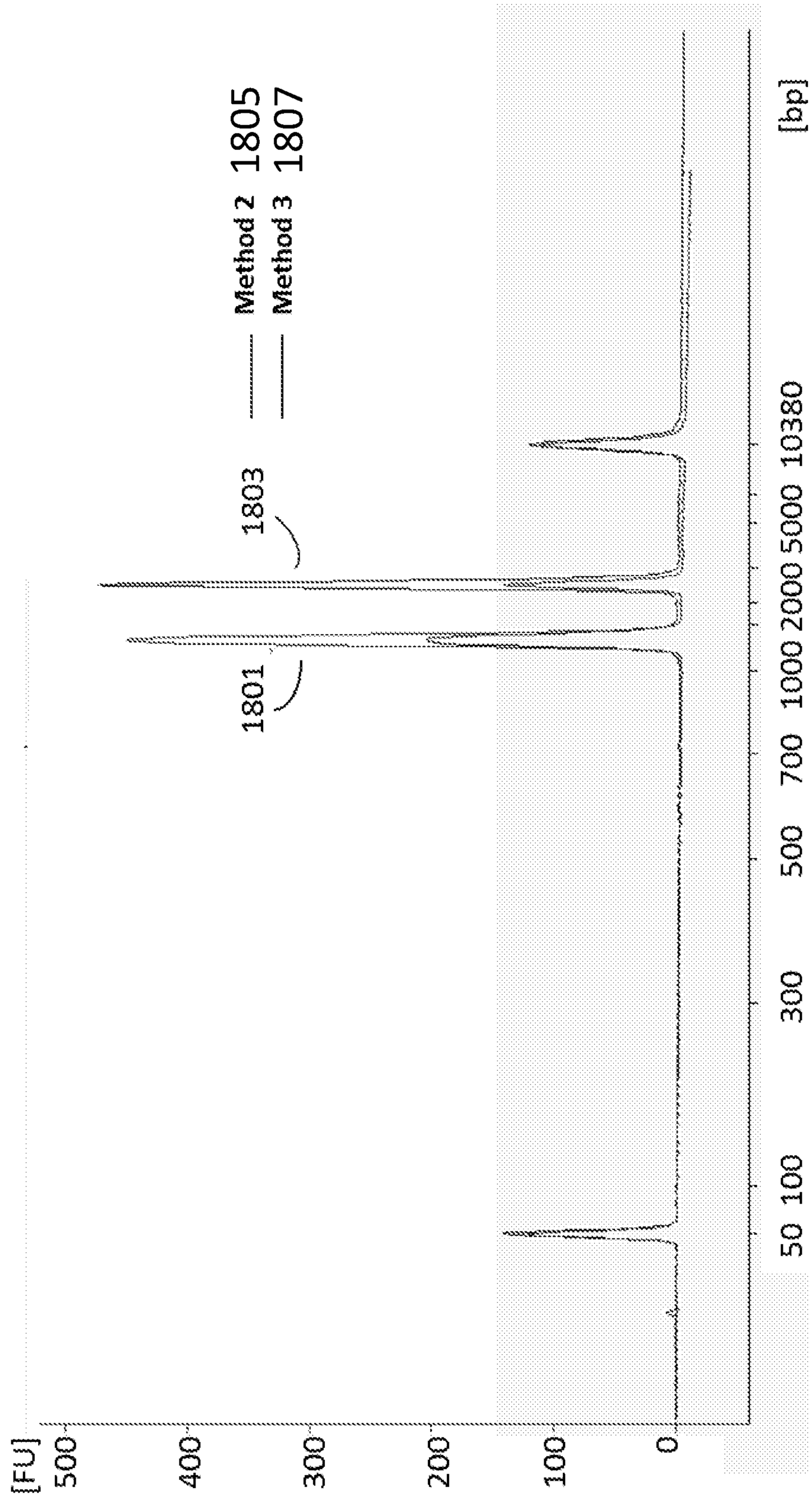

Method 2 1805
Method 3 1807
1803
1801
[bp]
50 100 300 500 700 1000 2000 5000 10380
[FU]
500 400 300 200 100 0

FIG. 18A

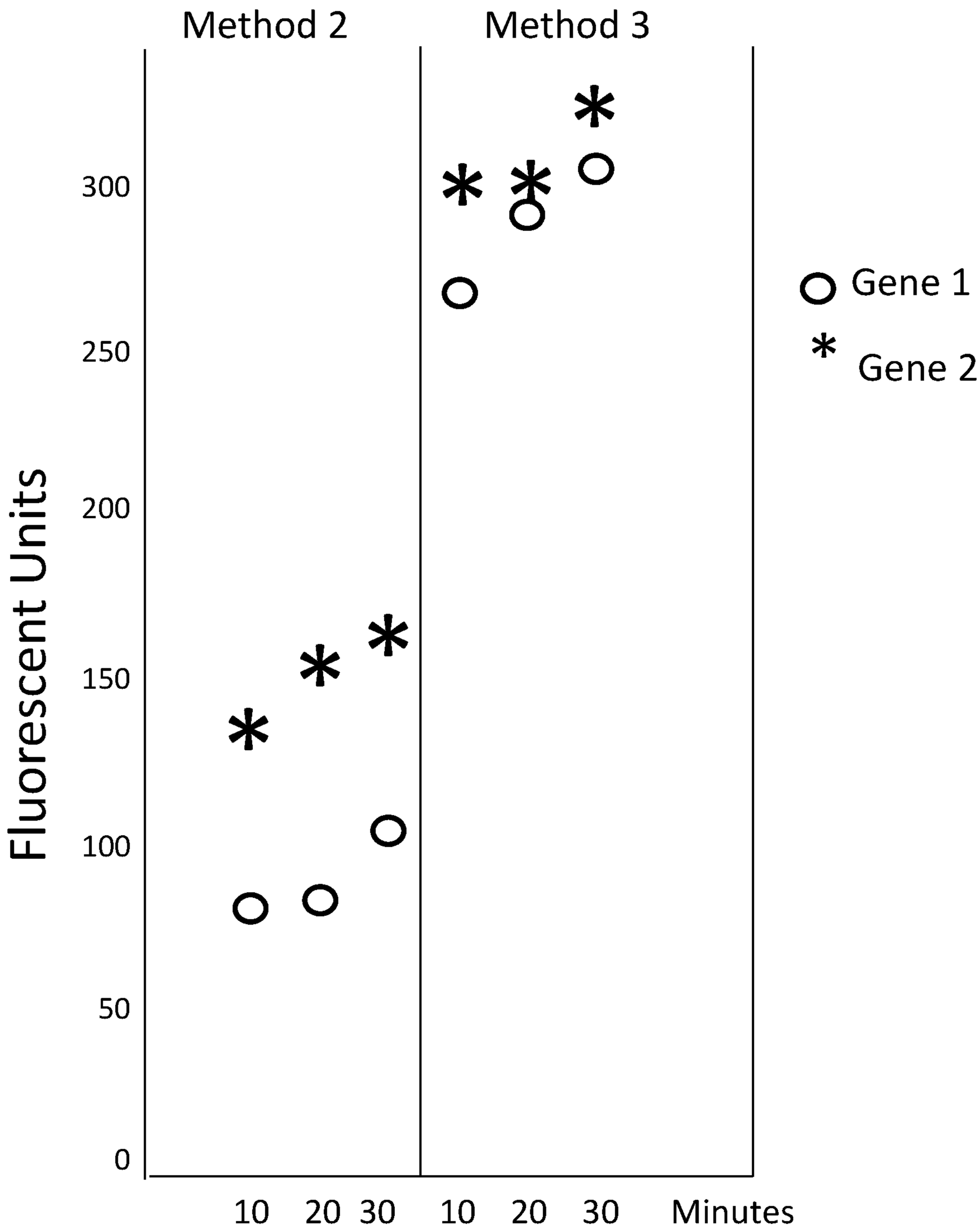

Method 2
Method 3
Fluorescent Units
300
250
200
150
100
50
0
Gene 1
Gene 2
10 20 30 10 20 30 Minutes

FIG. 18B

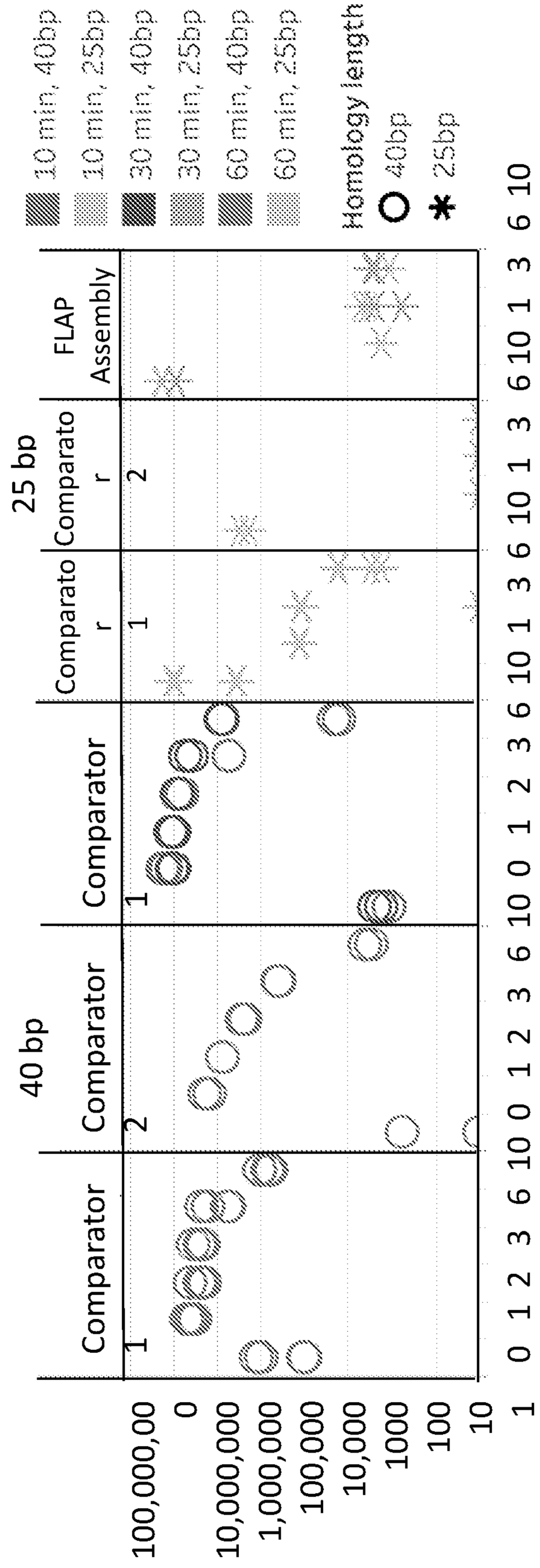

10 min, 40bp
10 min, 25bp
30 min, 40bp
30 min, 25bp
60 min, 40bp
60 min, 25bp
Homology length
40bp
25bp
25 bp
40 bp
Comparator 1
Comparator 2
FLAP Assembly
0 1 2 3 6 10 0 1 2 3 6 10 0 1 2 3 6 10 0 1 2 3 6 10
Fragments
CFUs (log scale)
100,000,000
10,000,000
1,000,000
100,000
10,000
1000
100
10
1
0

FIG. 19A

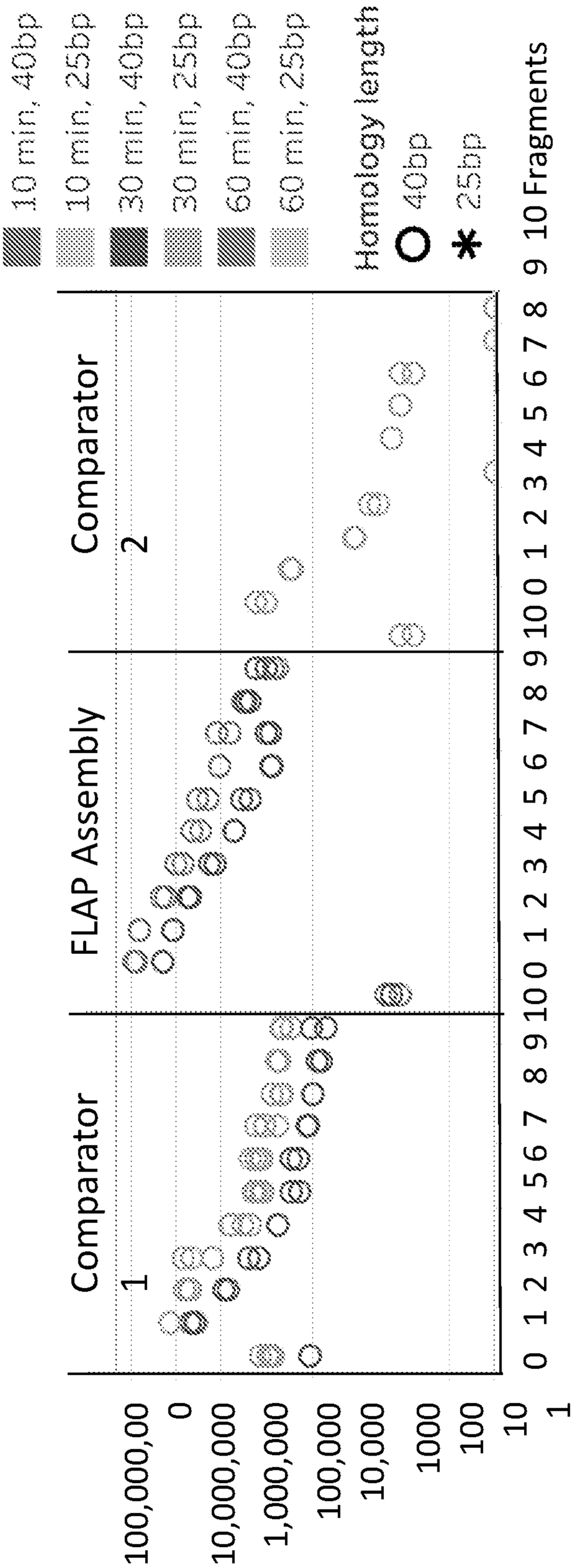

10 min, 40bp
10 min, 25bp
30 min, 40bp
30 min, 25bp
60 min, 40bp
60 min, 25bp
Homology length
40bp
25bp
1 2 3 4 5 6 7 8 9 10 Fragments
Comparator
2
FLAP Assembly
Comparator
1
0 1 2 3 4 5 6 7 8 9 10 0 1 2 3 4 5 6 7 8
100,000,00
0
10,000,000
1,000,000
100,000
10,000
1000
100
10
1
CFUs (log scale)

FIG. 19B

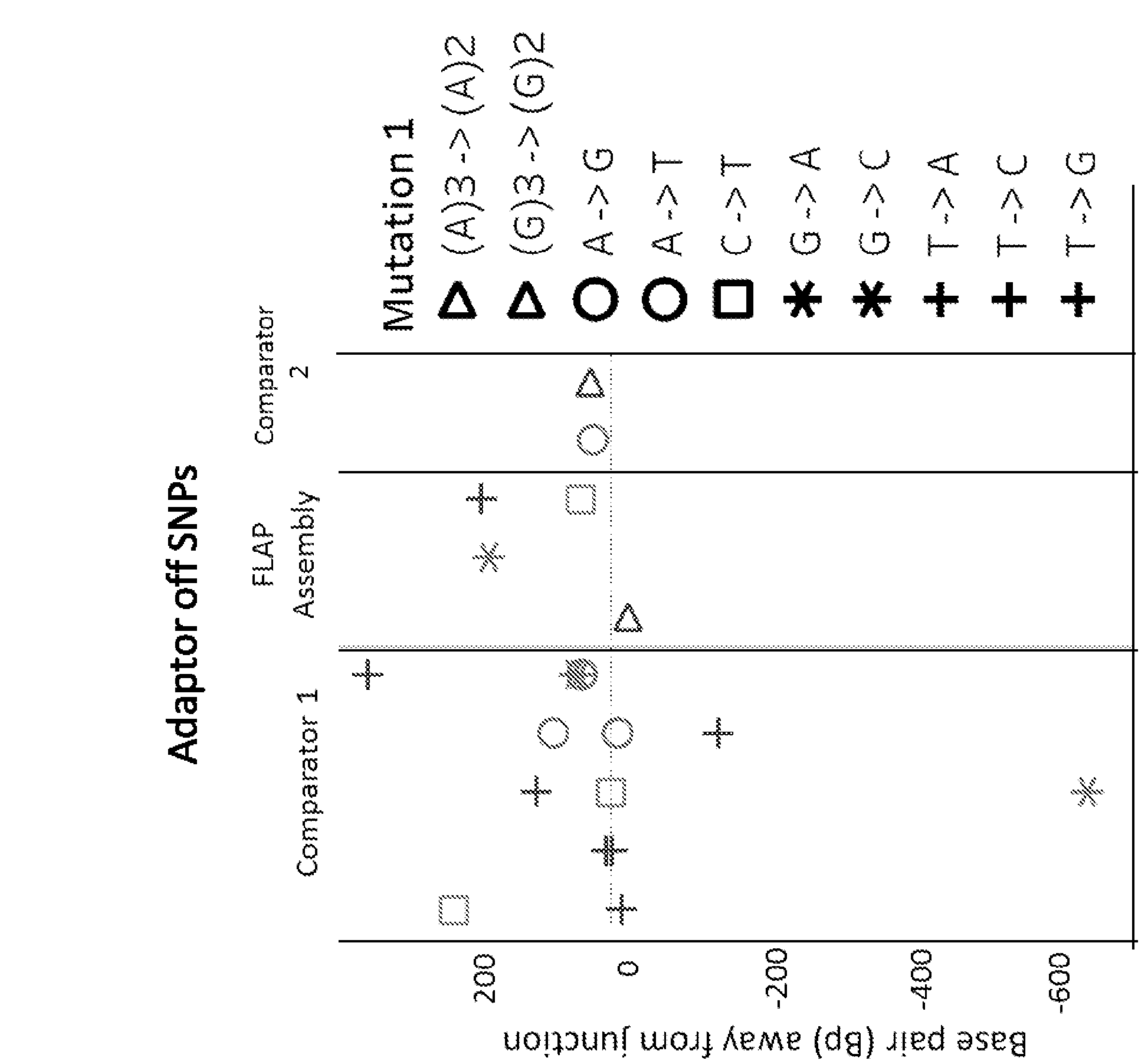

Adaptor off SNPs
Comparator 2
FLAP Assembly
Comparator 1
Mutation 1
(A)3 -> (A)2
(G)3 -> (G)2
A -> G
A -> T
C -> T
G -> A
G -> C
T -> A
T -> C
T -> G
200
0
-200
-400
-600
1 2 3 6 10 2 3 10
Base pair (Bp) away from junction

FIG. 20A

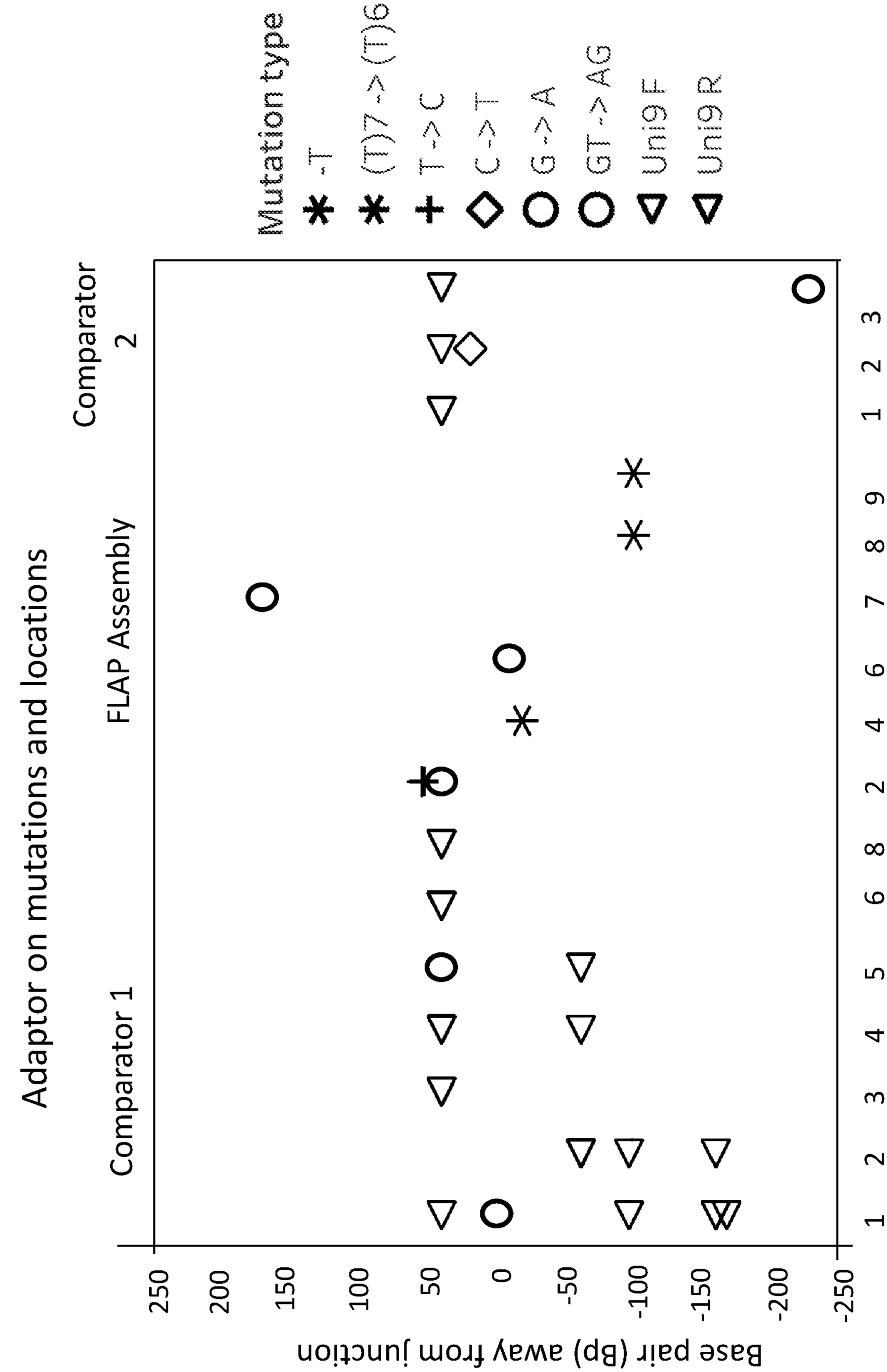

Mutation type
-T
(T)7 -> (T)6
T -> C
C -> T
G -> A
GT -> AG
Uni9 F
Uni9 R
Comparator 2
FLAP Assembly
Adaptor on mutations and locations
Comparator 1
Base pair (Bp) away from junction
250
200
150
100
50
0
-50
-100
-150
-200
-250
1 2 3 4 5 6 7 8 9 1 2 3 4 5 6 8 2 3
Number of Fragments

FIG. 20B

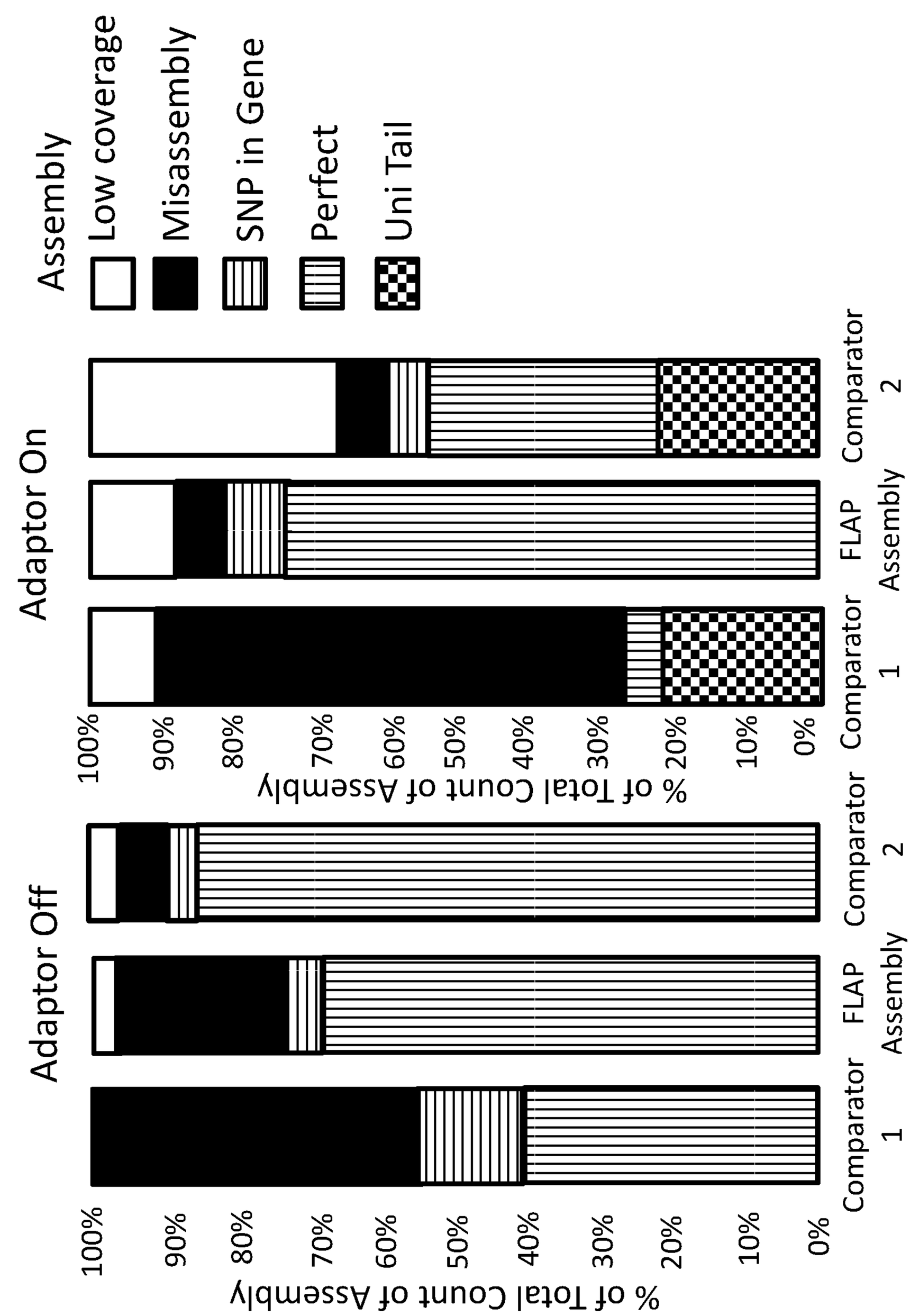

Assembly
Low coverage
Misassembly
SNP in Gene
Perfect
Uni Tail
Adaptor On
% of Total Count of Assembly
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
Comparator 2
FLAP Comparator 1 Assembly
Adaptor Off
% of Total Count of Assembly
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
FLAP Comparator 2 Assembly
Comparator 1

FIG. 20C

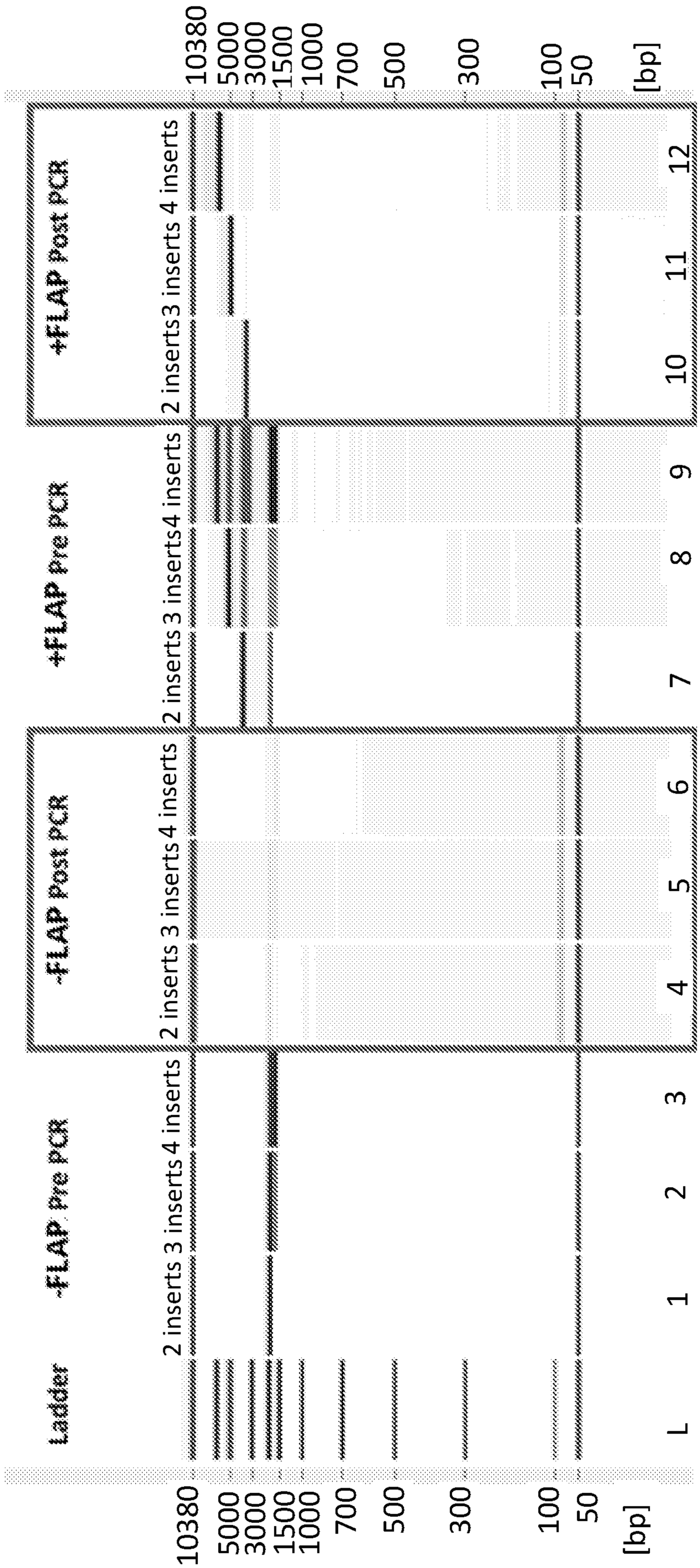

Ladder
-FLAP Pre PCR
2 inserts 3 inserts 4 inserts
-FLAP Post PCR
2 inserts 3 inserts 4 inserts
+FLAP Pre PCR
2 inserts 3 inserts 4 inserts
+FLAP Post PCR
2 inserts 3 inserts 4 inserts
L 1 2 3 4 5 6 7 8 9 10 11 12
[bp]
10380
5000
3000
1500
1000
700
500
300
100
50

FIG. 21A

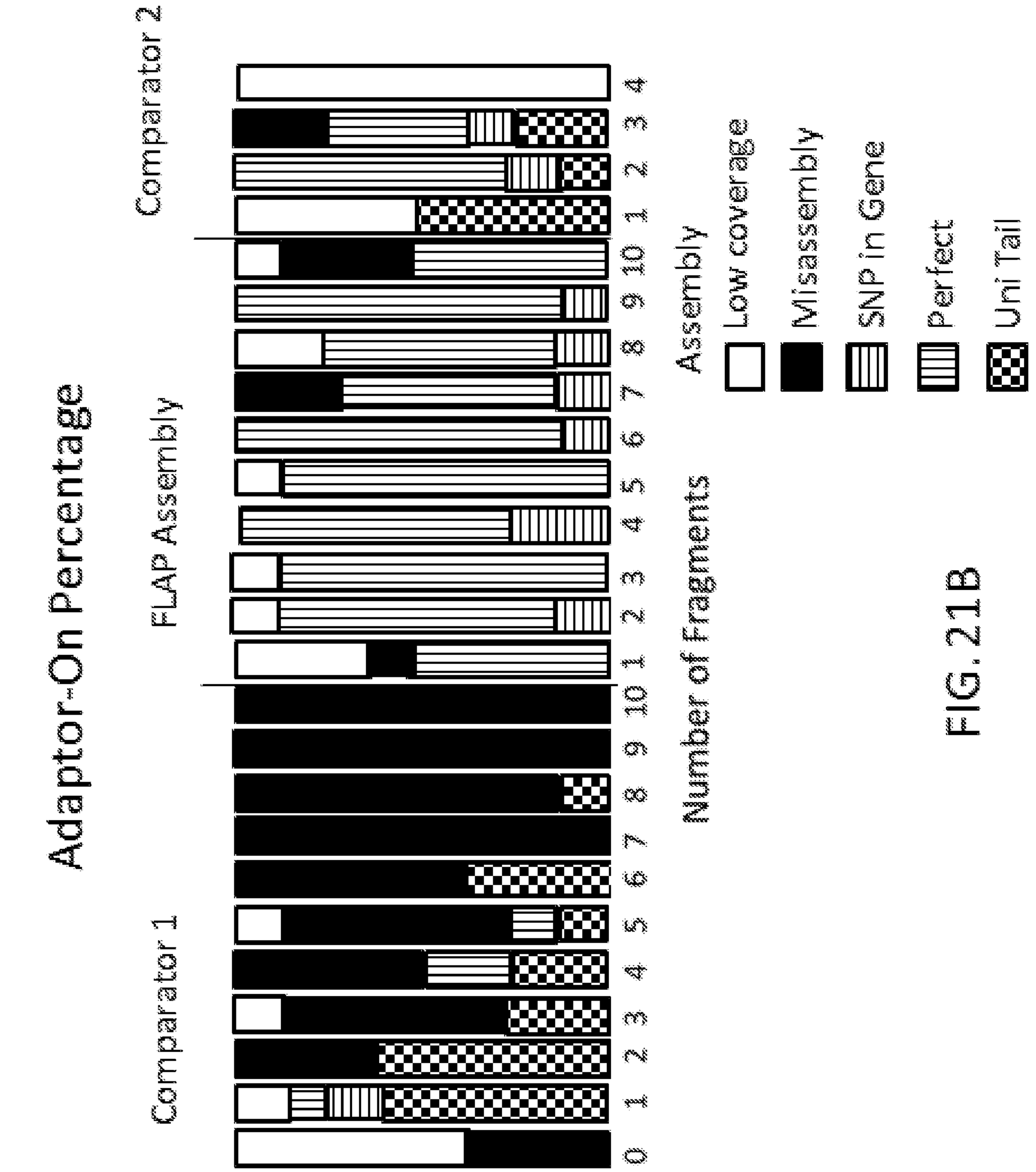

Adaptor-On Percentage
FLAP Assembly
Comparator 2
Comparator 1
Assembly
Low coverage
Misassembly
SNP in Gene
Perfect
Uni Tail
Number of Fragments
% of Total Count of Assembly
100%
80%
60%
40%
20%
0%
0
1 2 3 4 5 6 7 8 9 10 1 2 3

FIG. 21B

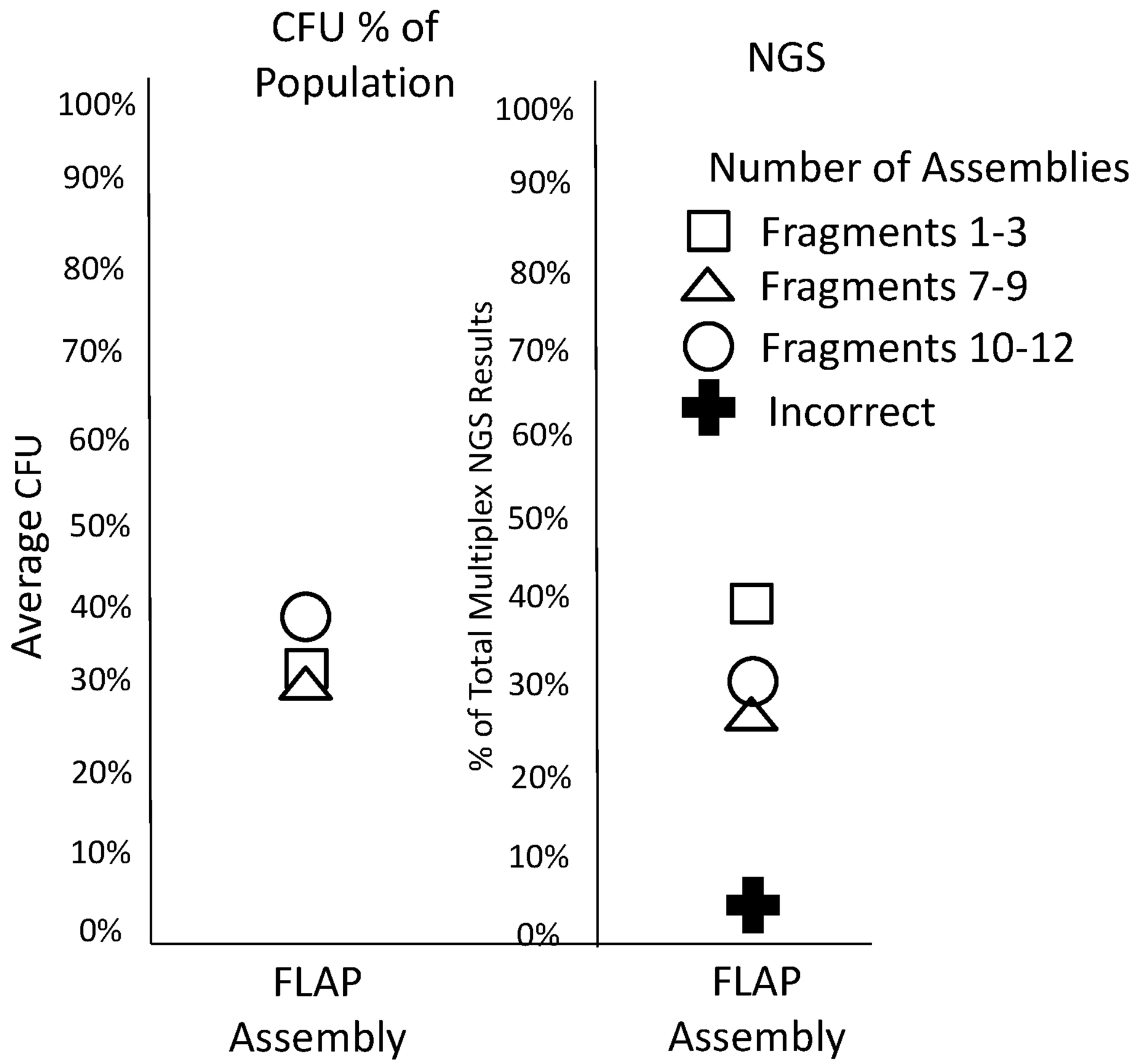

CFU % of Population
Average CFU
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
FLAP Assembly
NGS
% of Total Multiplex NGS Results
Number of Assemblies
Fragments 1-3
Fragments 7-9
Fragments 10-12
Incorrect
FLAP Assembly

FIG. 21C

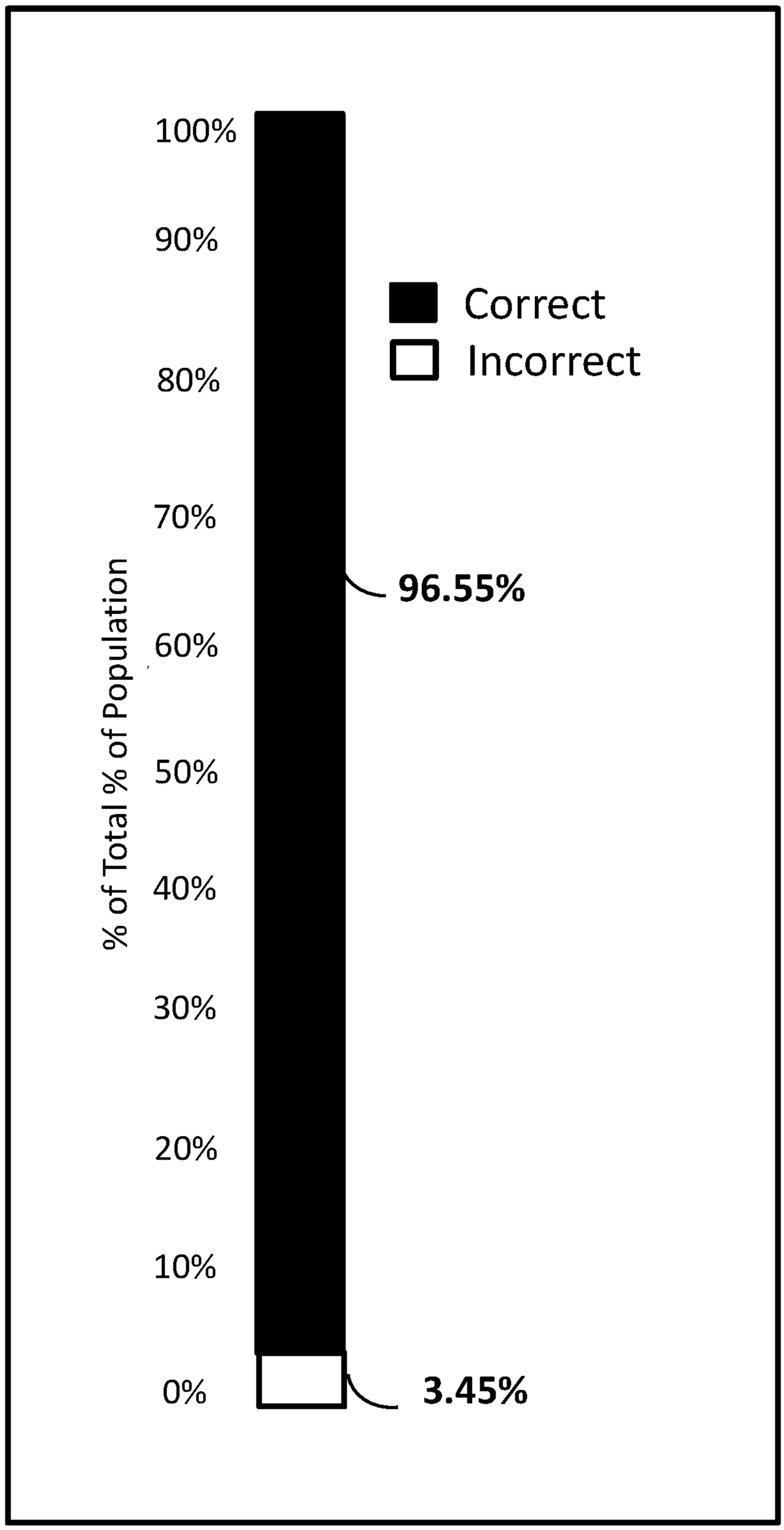

100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
% of Total % of Population
Correct
Incorrect
96.55%
3.45%

FIG. 21D

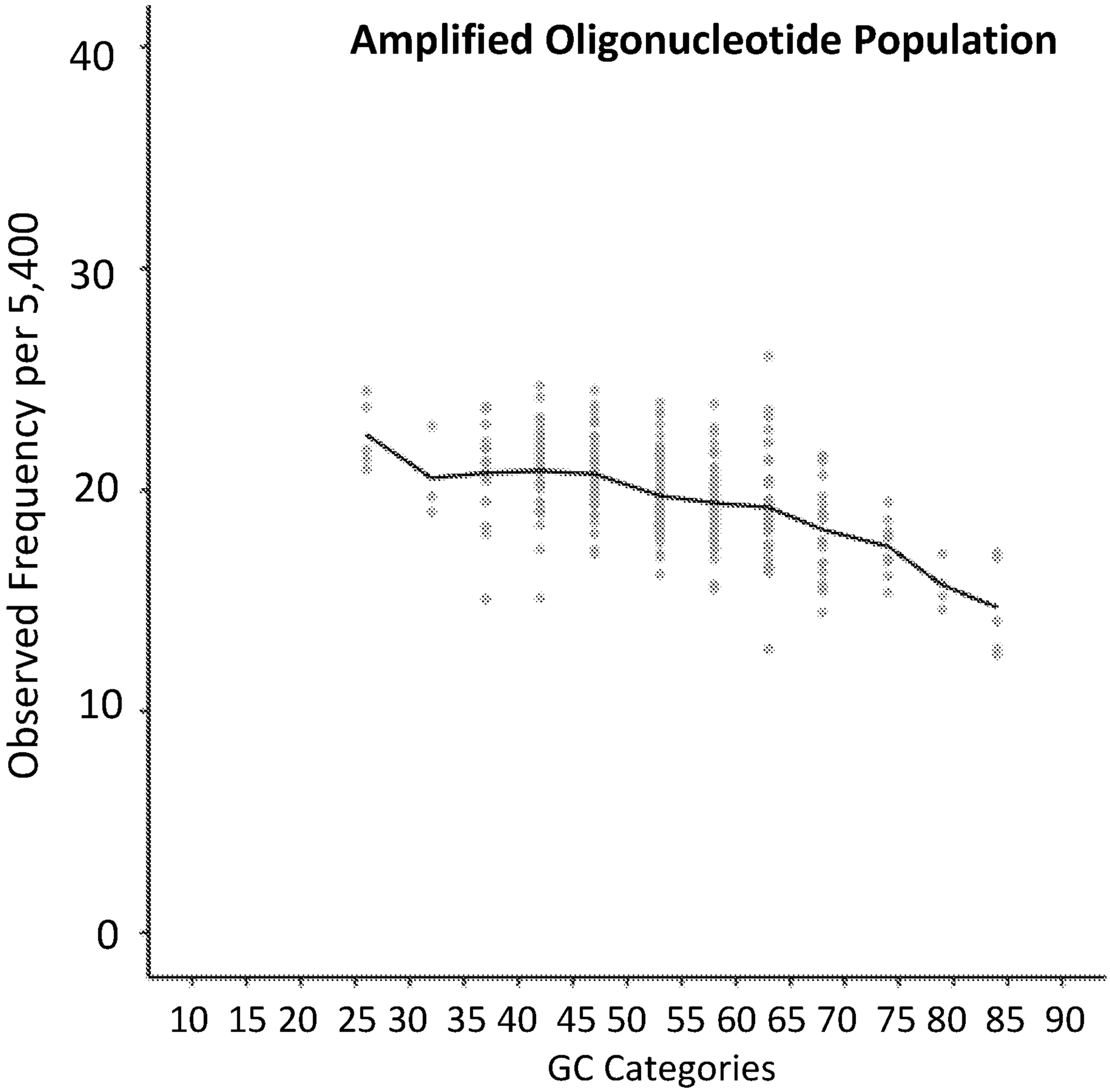

Amplified Oligonucleotide Population
Observed Frequency per 5,400
40
30
20
10
0
10 15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90
GC Categories

FIG. 22A

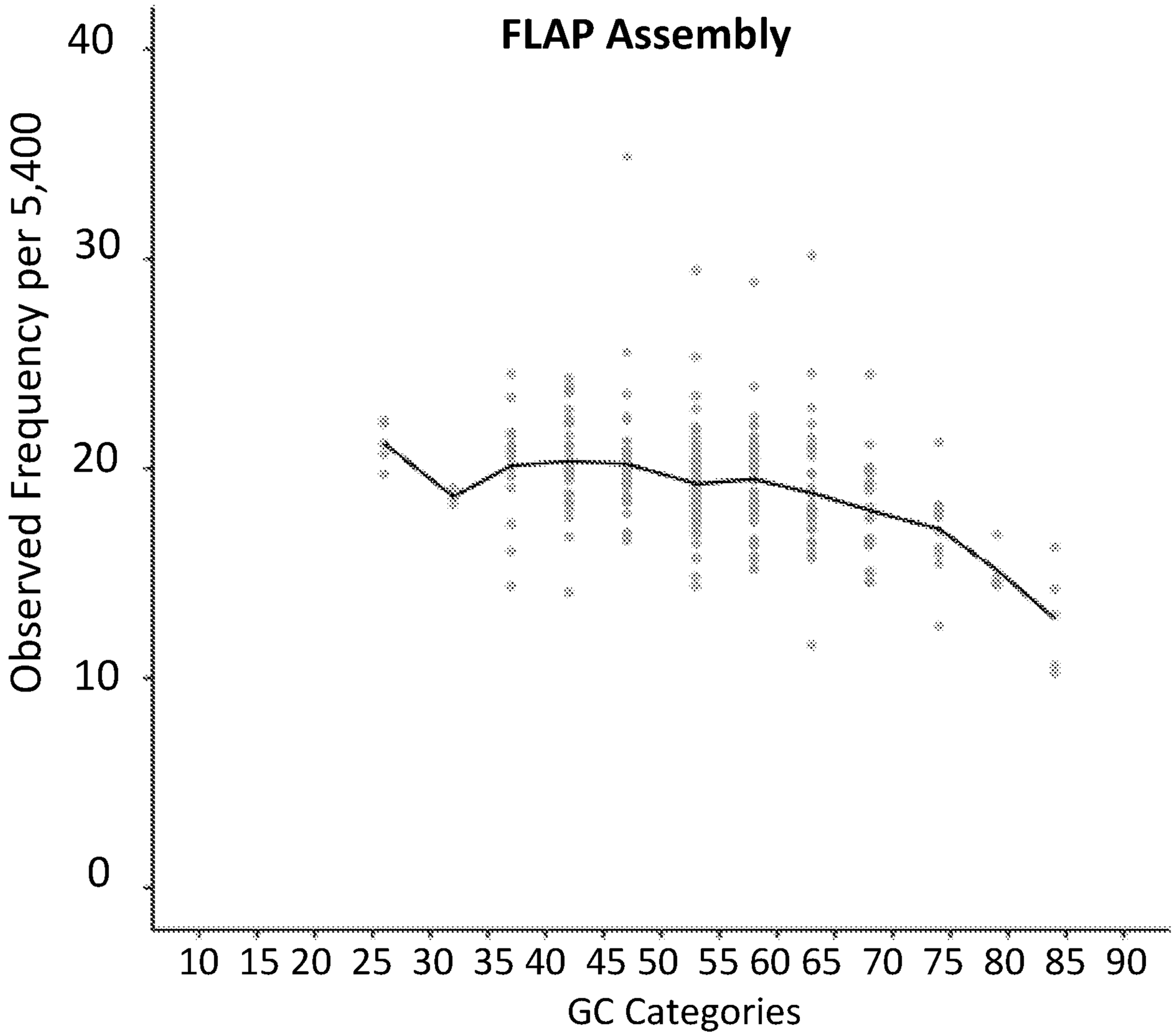

FLAP Assembly
Observed Frequency per 5,400
40
30
20
10
0
10 15 20 25 30 35 40 45 50 55 60 65 70 75 80 85 90
GC Categories

FIG. 22B

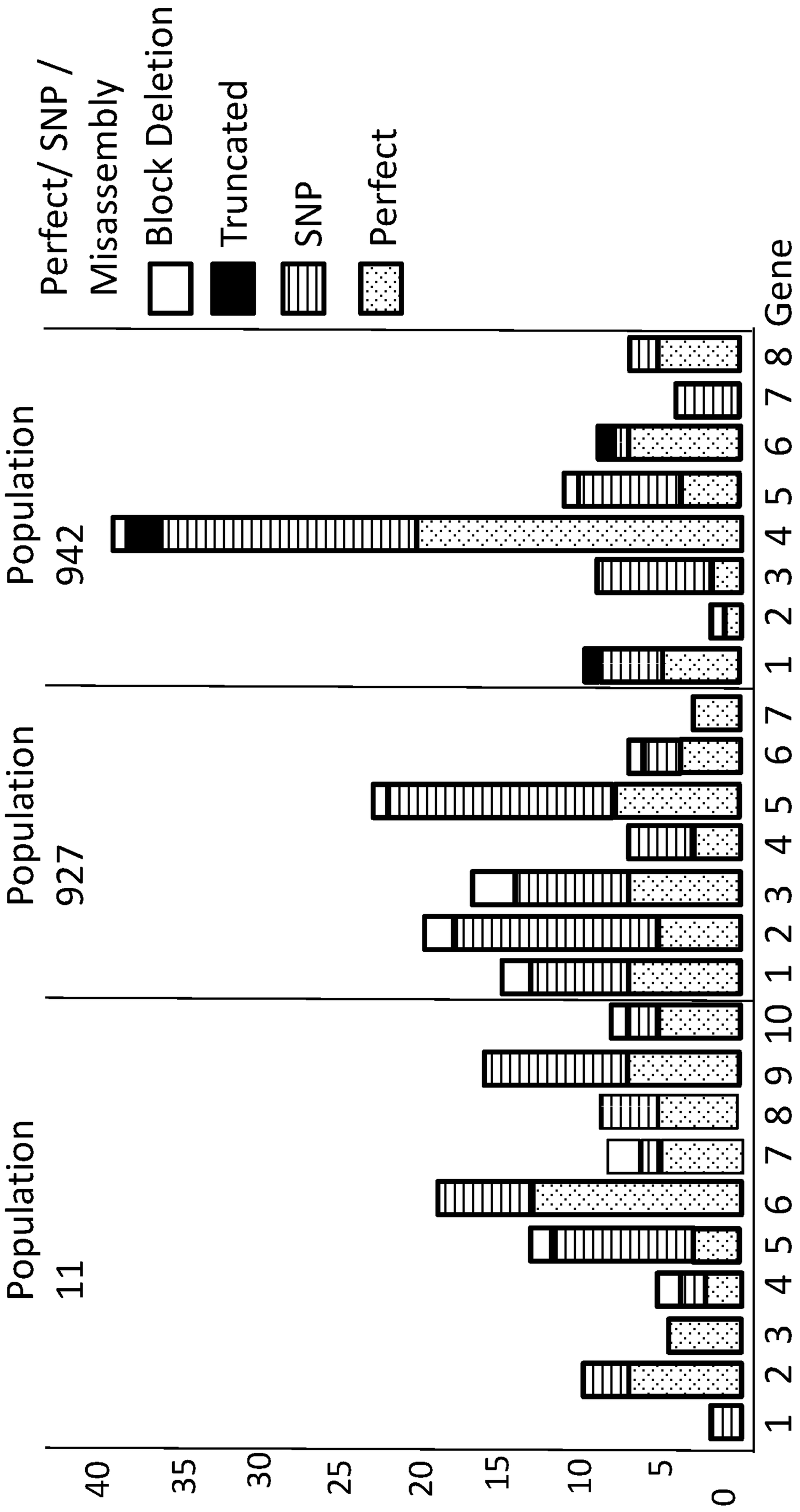

Perfect/ SNP / Misassembly
Block Deletion
Truncated
SNP
Perfect
Gene
Population 942
1 2 3 4 5 6 7 8
Population 927
1 2 3 4 5 6 7
Population 11
1 2 3 4 5 6 7 8 9 10
Count of Genes
0 5 10 15 20 25 30 35 40

FIG. 23