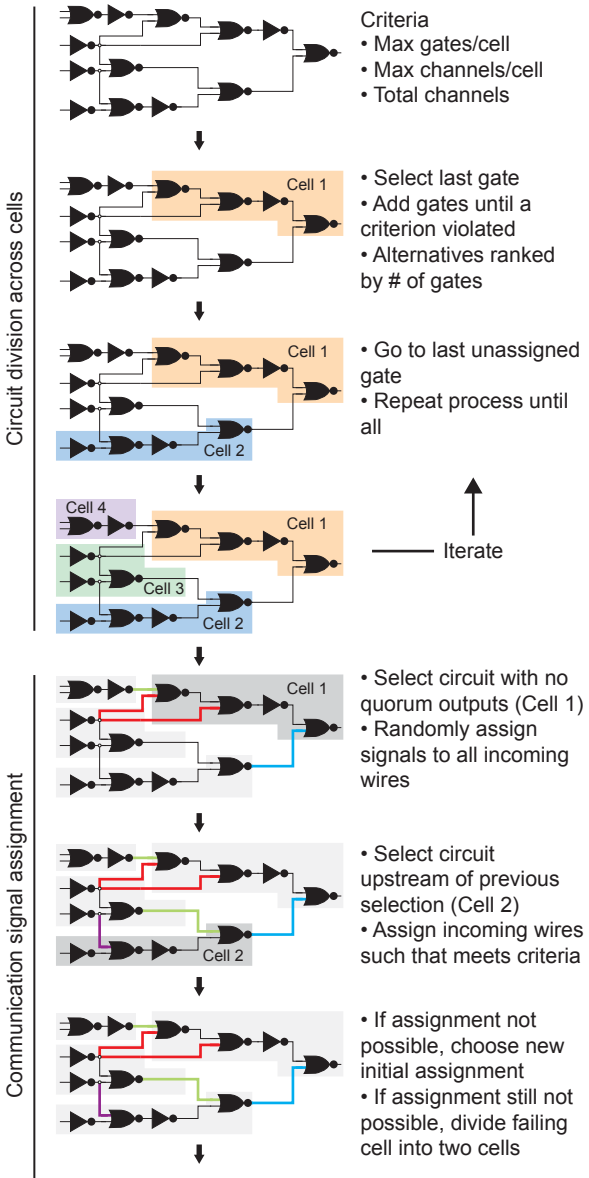

Circuit division across cells
Criteria
• Max gates/cell
• Max channels/cell
• Total channels
Cell 1
• Select last gate
• Add gates until a criterion violated
• Alternatives ranked by # of gates
Cell 1
Cell 2
• Go to last unassigned gate
• Repeat process until all
Cell 4
Cell 1
Cell 3
Cell 2
Iterate
Communication signal assignment
Cell 1
• Select circuit with no quorum outputs (Cell 1)
• Randomly assign signals to all incoming wires
Cell 2
• Select circuit upstream of previous selection (Cell 2)
• Assign incoming wires such that meets criteria
• If assignment not possible, choose new initial assignment
• If assignment still not possible, divide failing cell into two cells

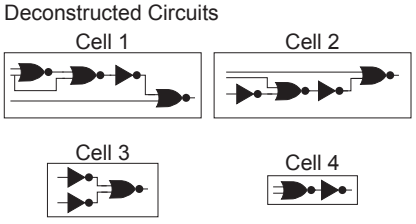

Deconstructed Circuits
Cell 1
Cell 2
Cell 3
Cell 4

Figure 2

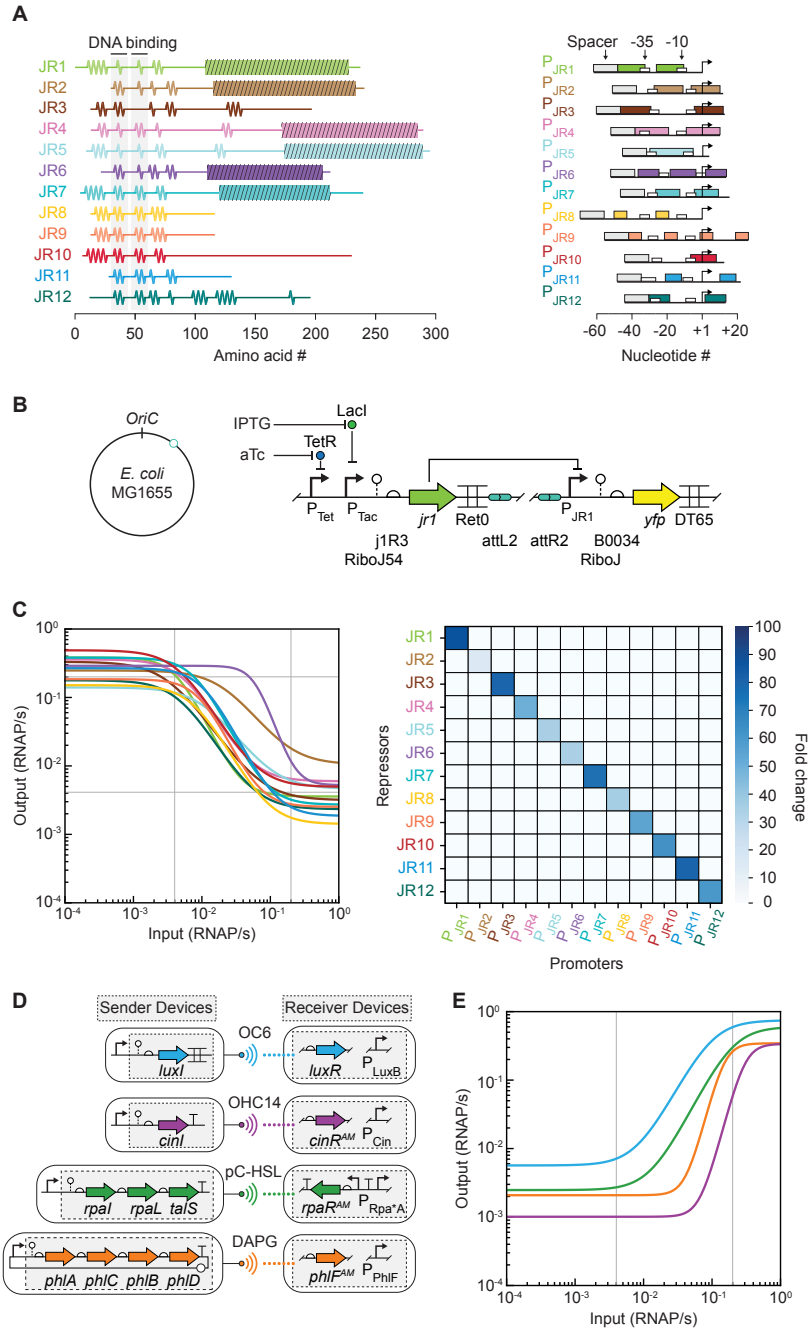

A
DNA binding
JR1
JR2
JR3
JR4
JR5
JR6
JR7
JR8
JR9
JR10
JR11
JR12
0
50
100
150
200
250
300
Amino acid #
Spacer
-35
-10
-60
-40
-20
+1
+20
Nucleotide #
B
OriC
E. coli
MG1655
IPTG
LacI
aTc
TetR
P Tet
P Tac
jr1
Ret0
P JR1
yfp
DT65
j1R3
RiboJ54
attL2
attR2
B0034
RiboJ
C
Output (RNAP/s)
Input (RNAP/s)
Repressors
Promoters
Fold change
D
Sender Devices
Receiver Devices
luxI
OC6
luxR
P LuxB
cinI
OHC14
cinR^AM
P Cin
rpaI rpaL talS
pC-HSL
rpaR^AM
P Rpa*A
phlA phlC phlB phlD
DAPG
phlF^AM
P PhlF
E
Output (RNAP/s)
Input (RNAP/s)

Figure 3

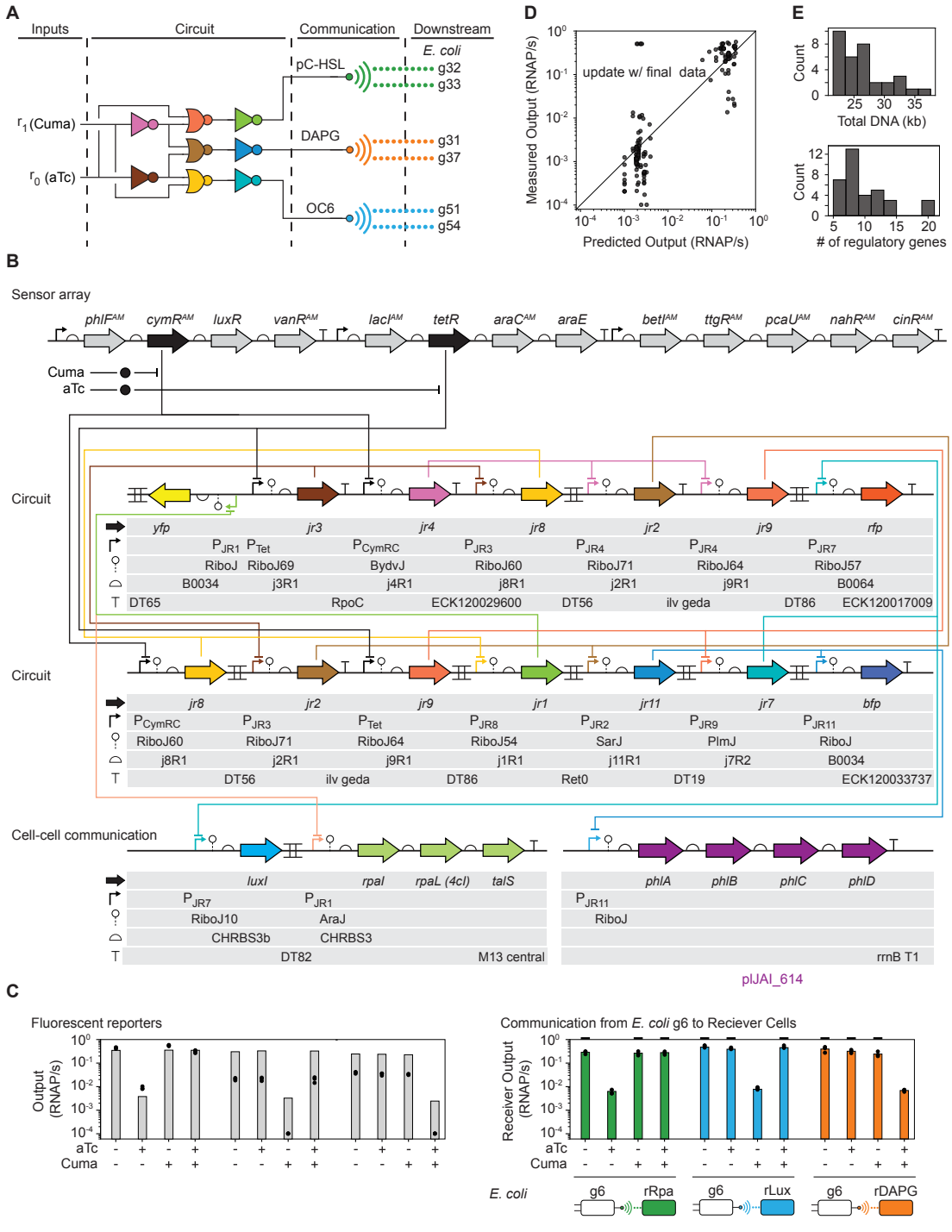
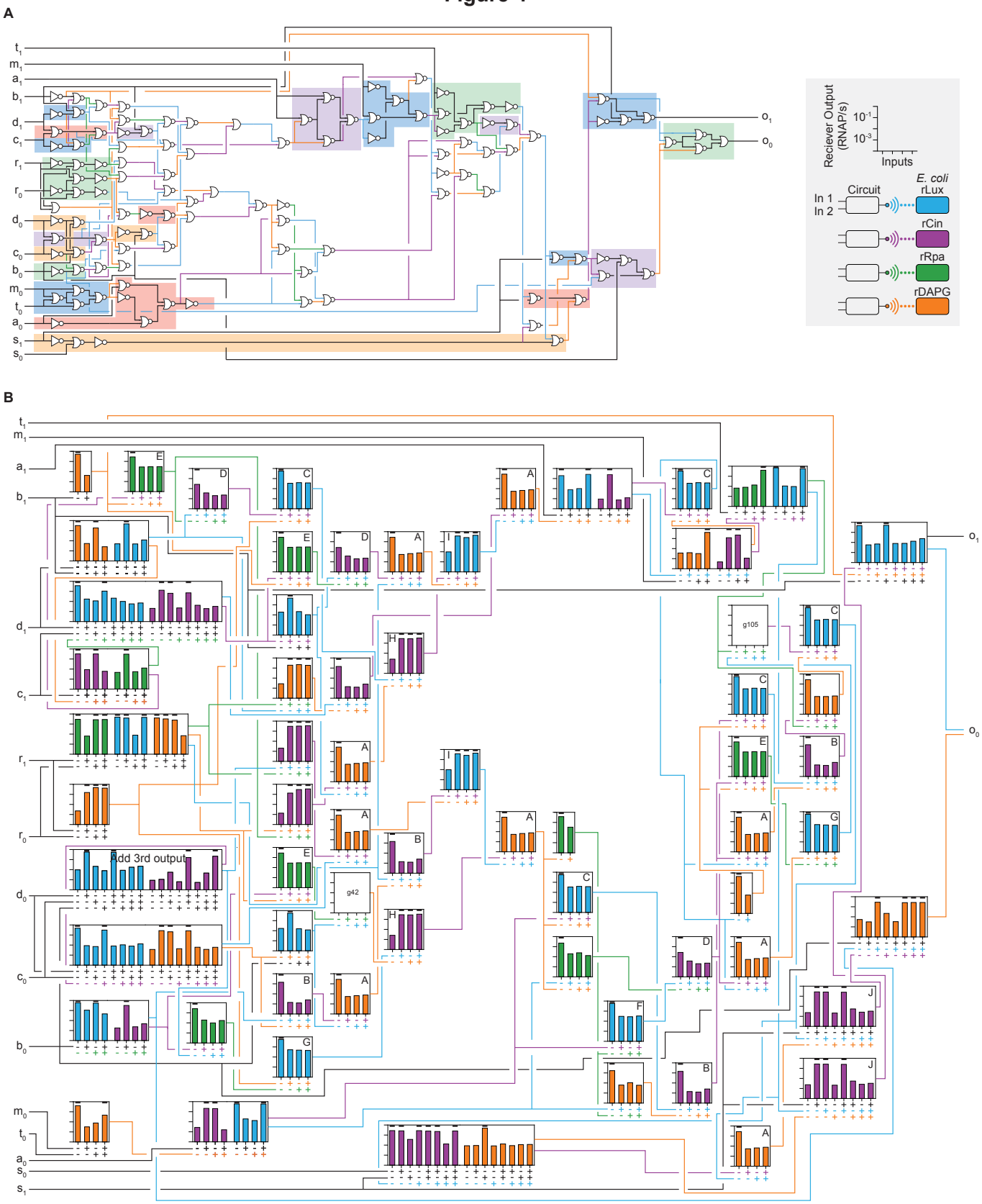

A
Inputs
Circuit
Communication
Downstream
E. coli
pC-HSL
g32
g33
r1 (Cuma)
DAPG
g31
g37
r0 (aTc)
OC6
g51
g54
D
update w/ final data
Measured Output (RNAP/s)
Predicted Output (RNAP/s)
E
Count
Total DNA (kb)
# of regulatory genes
B
Sensor array
phlF^AM cymR^AM luxR vanR^AM lacI^AM tetR araC^AM araE betI^AM ttgR^AM pcaU^AM nahR^AM cinR^AM
Cuma
aTc
Circuit
yfp jr3 jr4 jr8 jr2 jr9 rfp
P_JR1 P_Tet P_CymRC P_JR3 P_JR4 P_JR4 P_JR7
RiboJ RiboJ69 BydvJ RiboJ60 RiboJ71 RiboJ64 RiboJ57
B0034 j3R1 j4R1 j8R1 j2R1 j9R1 B0064
DT65 RpoC ECK120029600 DT56 ilv geda DT86 ECK120017009
Circuit
jr8 jr2 jr9 jr1 jr11 jr7 bfp
P_CymRC P_JR3 P_Tet P_JR8 P_JR2 P_JR9 P_JR11
RiboJ60 RiboJ71 RiboJ64 RiboJ54 SarJ PlmJ RiboJ
j8R1 j2R1 j9R1 j1R1 j11R1 j7R2 B0034
DT56 ilv geda DT86 Ret0 DT19 ECK120033737
Cell-cell communication
luxI rpaI rpaL (4cl) talS
P_JR7 P_JR1
RiboJ10 AraJ
CHRBS3b CHRBS3
DT82 M13 central
phlA phlB phlC phlD
P_JR11
RiboJ
rrnB T1
pIJAI_614
C
Fluorescent reporters
Output (RNAP/s)
aTc
Cuma
Communication from E. coli g6 to Reciever Cells
Receiver Output (RNAP/s)
E. coli
g6 rRpa
g6 rLux
g6 rDAPG

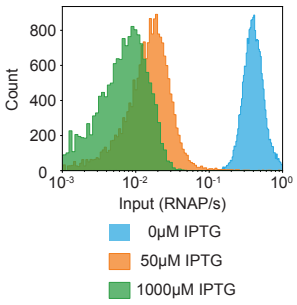
Figure 4

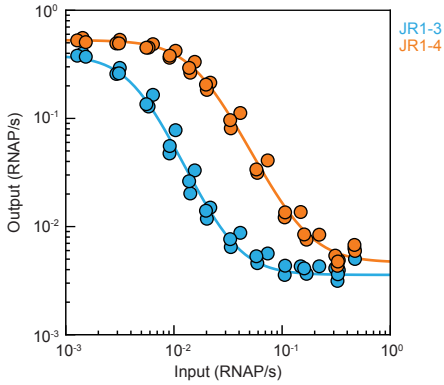

A
B
Reciever Output (RNAP/s)
Inputs
Circuit
In 1
In 2
E. coli
rLux
rCin
rRpa
rDAPG
Add 3rd output
g105
g42

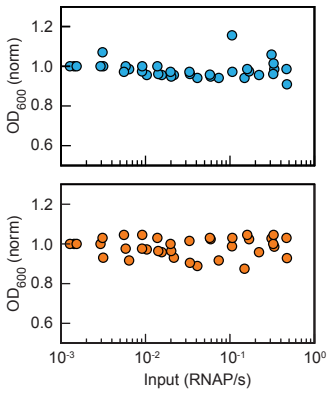
Gate JR1:pJR1

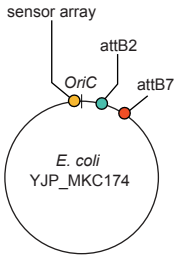
Source host/phage: E. coli/Lambda
Repressor name (protein accession #): cI (CAB96428.1)[citation]
Genome encoded NOT gates: JAI_MKC201, JAI_MKC202
Plasmid encoded NOT gates: JAI_DHC179, JAI_DHC180

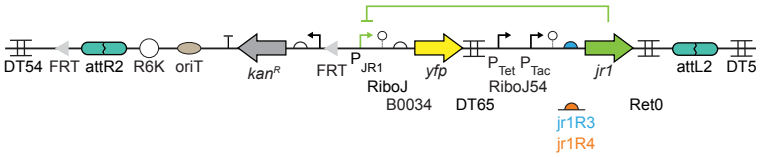
Genome encoded gate



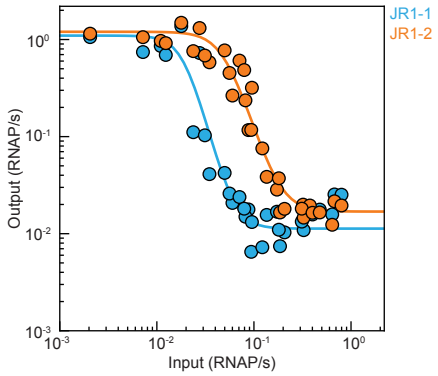


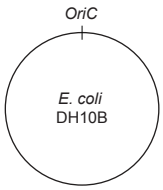






Plasmid encoded gate

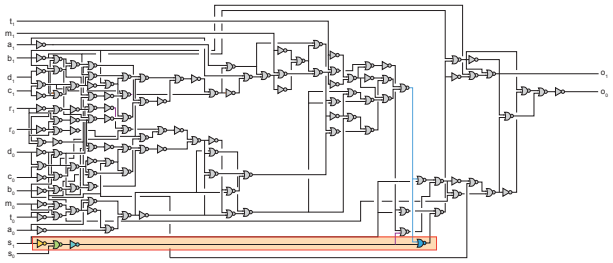




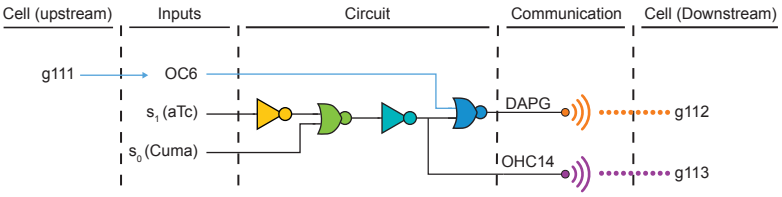
Circuit g101

Strain: *E. coli* g101

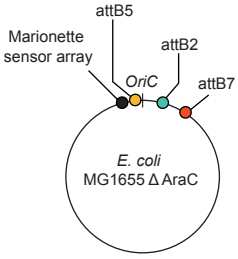
Amount of recombinant DNA: 32,213 bp

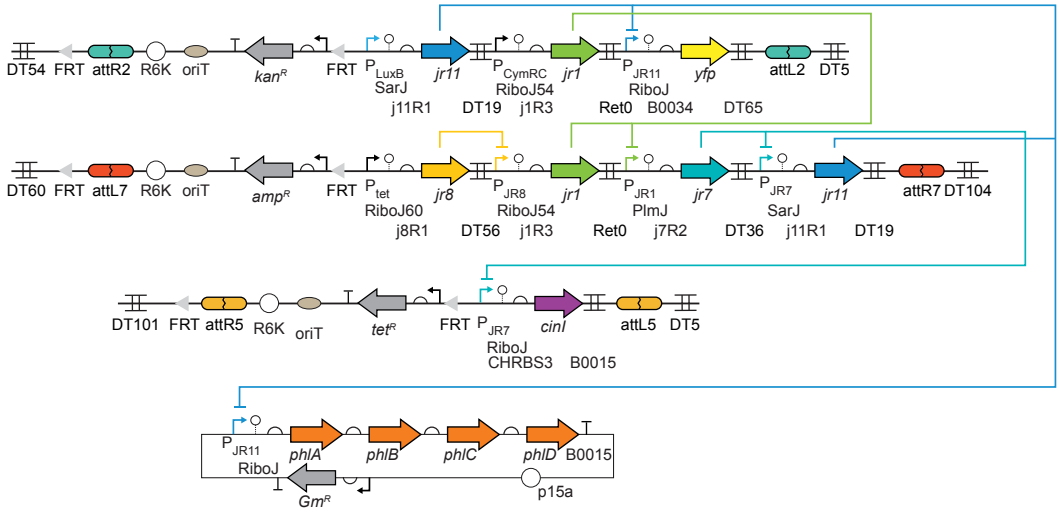


Circuit diagram



Genetic map





Circuit data