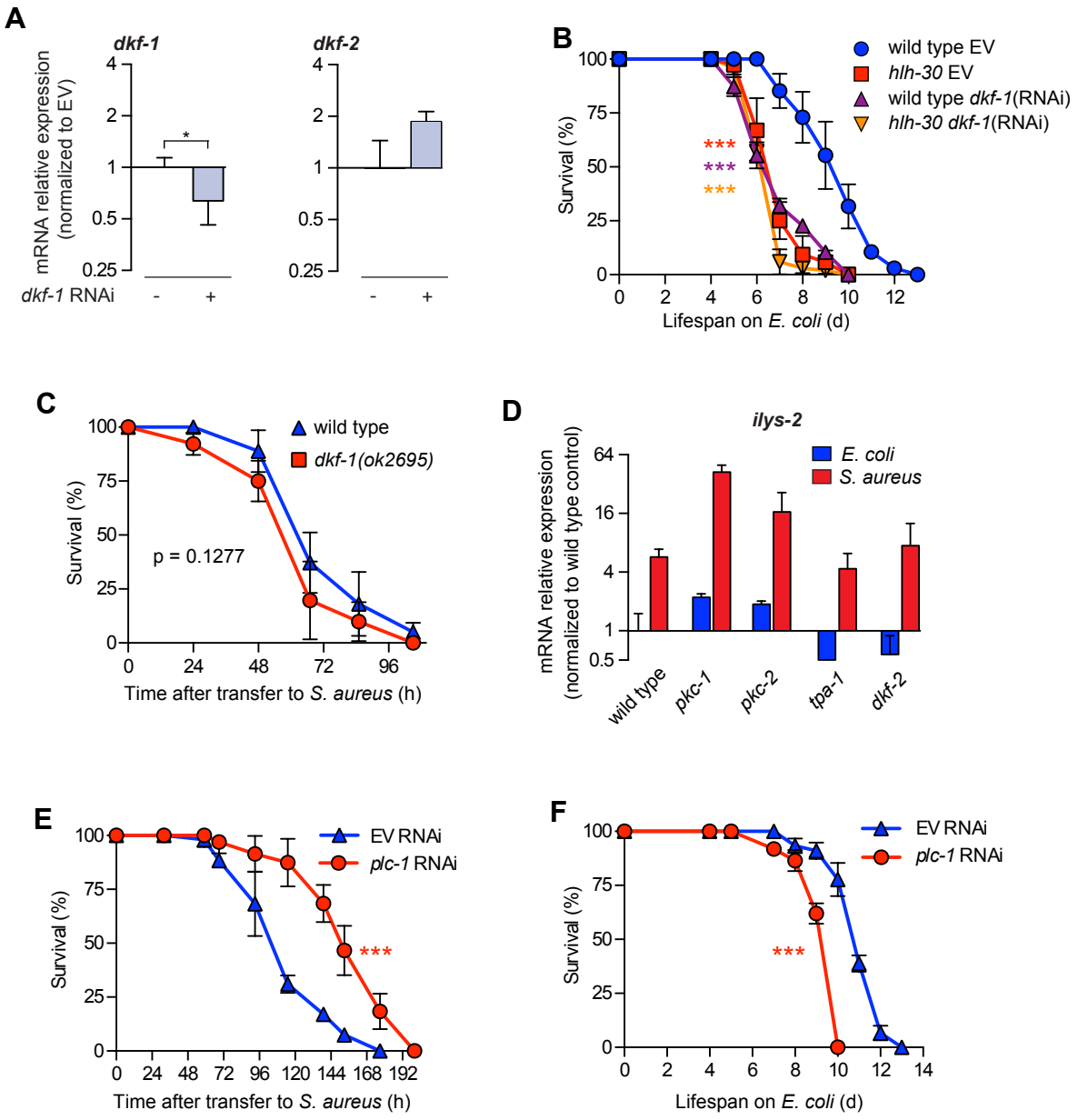
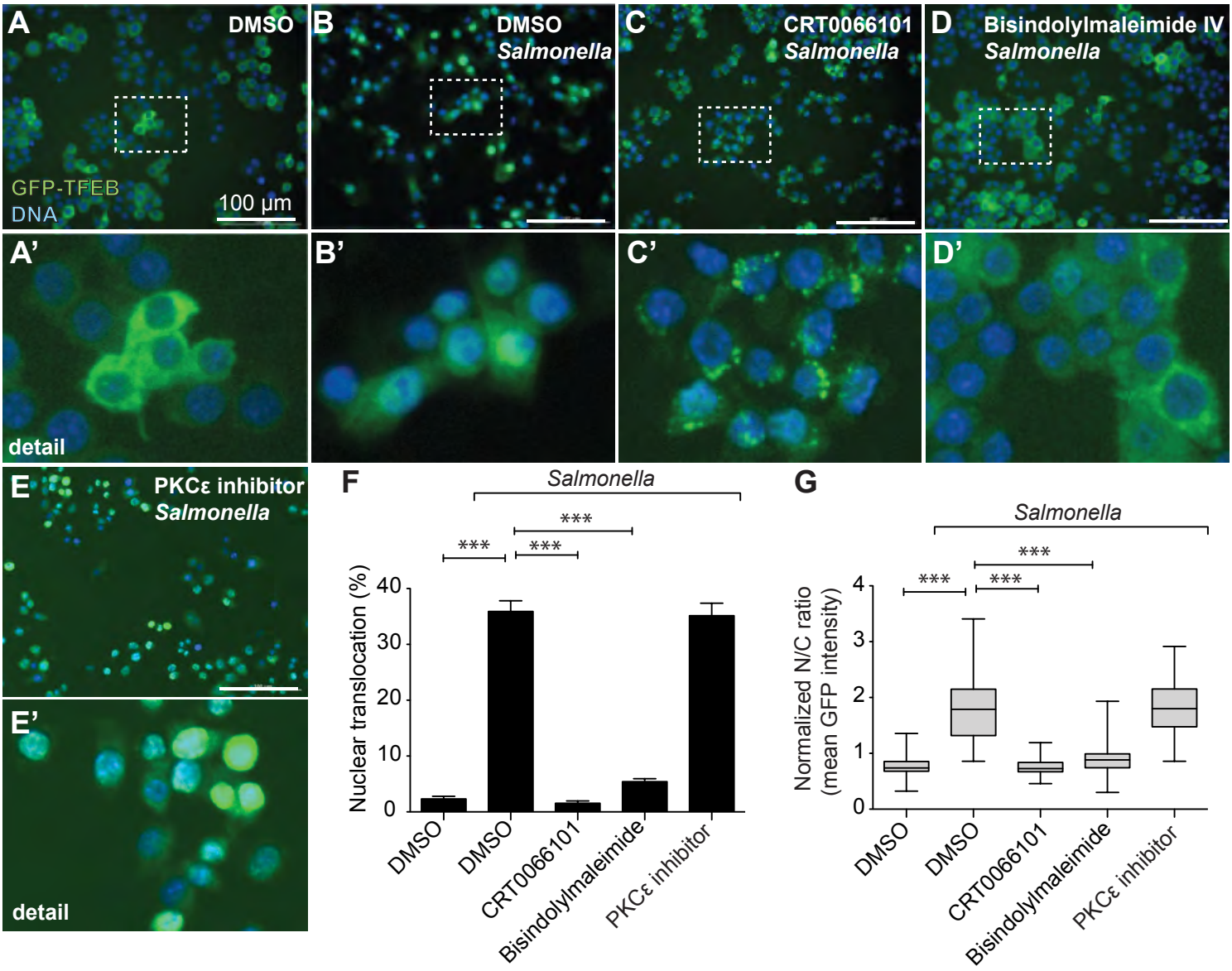
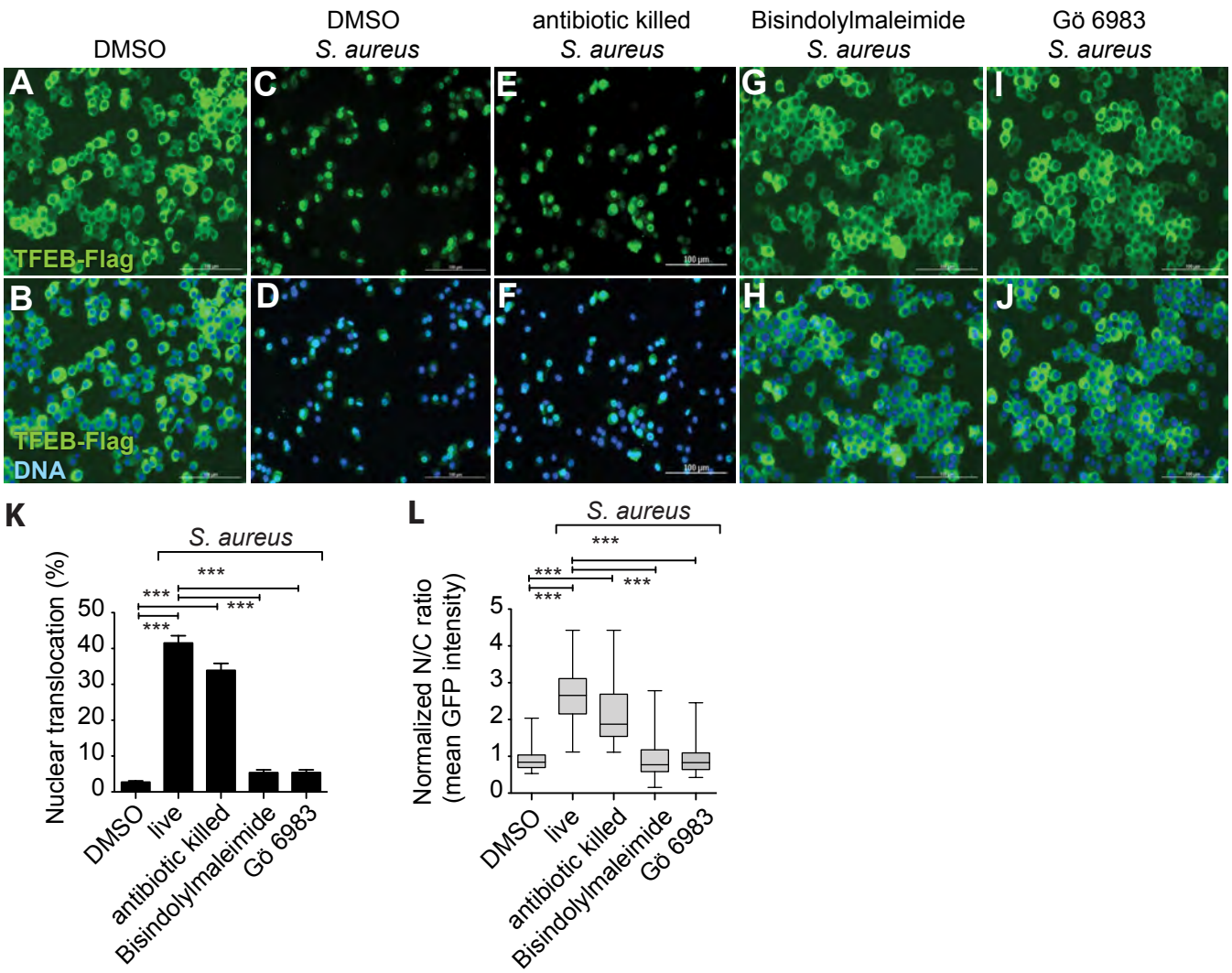

A
dkf-1
dkf-2
mRNA relative expression (normalized to EV)
4
2
1
0.5
0.25
*
dkf-1 RNAi
-
+
B
wild type EV
hlh-30 EV
wild type dkf-1(RNAi)
hlh-30 dkf-1(RNAi)
Survival (%)
100
75
50
25
0
***
***
***
0
2
4
6
8
10
12
Lifespan on E. coli (d)
C
wild type
dkf-1(ok2695)
Survival (%)
p = 0.1277
0
24
48
72
96
Time after transfer to S. aureus (h)
D
ilys-2
E. coli
S. aureus
mRNA relative expression (normalized to wild type control)
64
16
4
1
0.5
wild type
pkc-1
pkc-2
tpa-1
dkf-2
E
EV RNAi
plc-1 RNAi
Survival (%)
***
0
24
48
72
96
120
144
168
192
Time after transfer to S. aureus (h)
F
EV RNAi
plc-1 RNAi
Survival (%)
***
0
2
4
6
8
10
12
14
Lifespan on E. coli (d)


A
DMSO
GFP-TFEB
DNA
100 µm
B
DMSO
Salmonella
C
CRT0066101
Salmonella
D
Bisindolylmaleimide IV
Salmonella
A'
detail
B'
C'
D'
E
PKCε inhibitor
Salmonella
E'
detail
F
Salmonella
***
***
***
Nuclear translocation (%)
40
30
20
10
0
DMSO
DMSO
CRT0066101
Bisindolylmaleimide
PKCε inhibitor
G
Salmonella
***
***
***
Normalized N/C ratio
(mean GFP intensity)
4
3
2
1
0
DMSO
DMSO
CRT0066101
Bisindolylmaleimide
PKCε inhibitor


DMSO
DMSO
S. aureus
antibiotic killed
S. aureus
Bisindolylmaleimide
S. aureus
Gö 6983
S. aureus
A
C
E
G
I
TFEB-Flag
B
D
F
H
J
TFEB-Flag
DNA
K
S. aureus
Nuclear translocation (%)
50
40
30
20
10
0
***
***
***
***
***
DMSO
live
antibiotic killed
Bisindolylmaleimide
Gö 6983
L
S. aureus
Normalized N/C ratio
(mean GFP intensity)
5
4
3
2
1
0
***
***
***
***
DMSO
live
antibiotic killed
Bisindolylmaleimide
Gö 6983

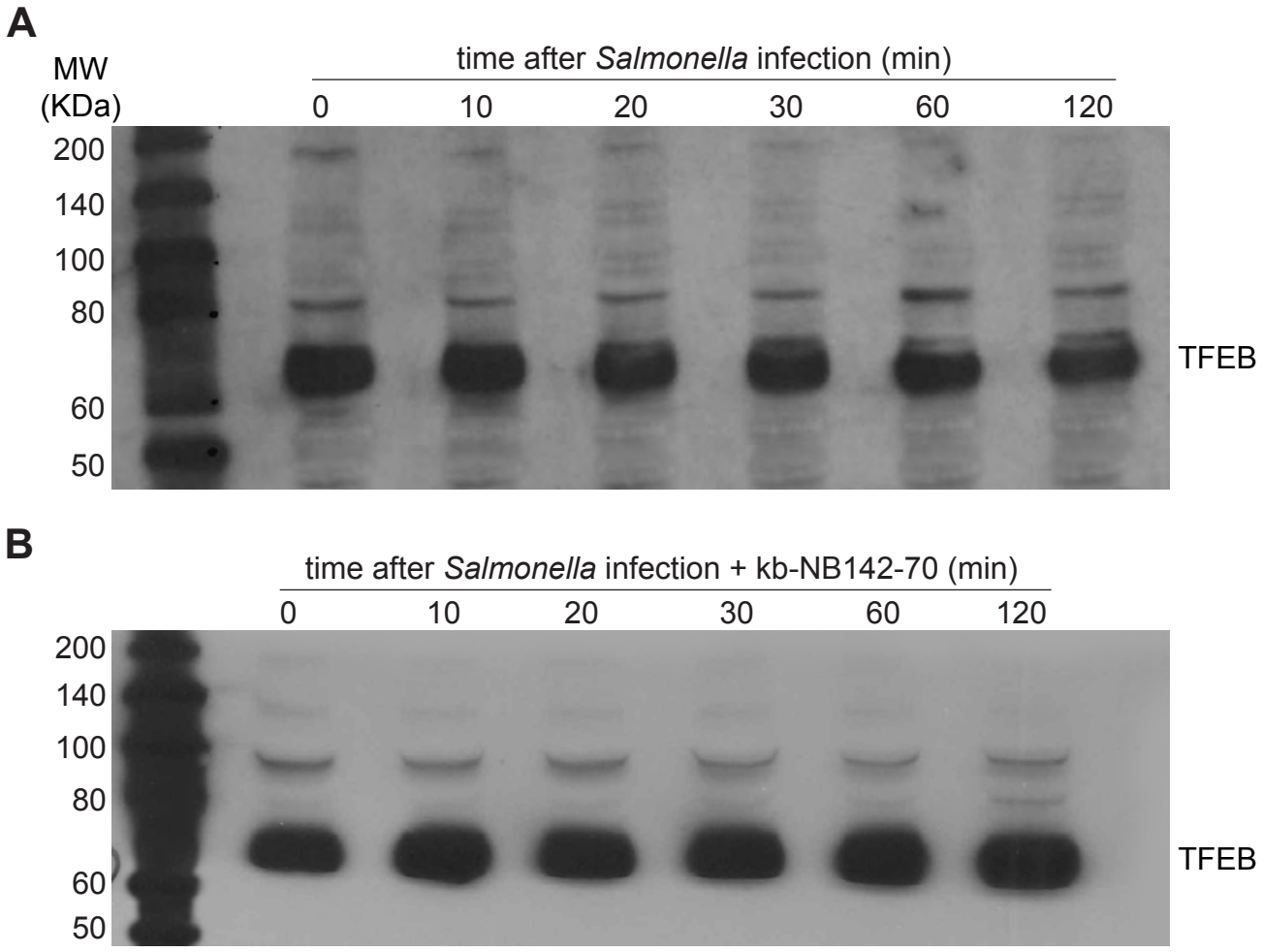

A
MW
(KDa)
time after Salmonella infection (min)
0
10
20
30
60
120
200
140
100
80
60
50
TFEB
B
time after Salmonella infection + kb-NB142-70 (min)
0
10
20
30
60
120
200
140
100
80
60
50
TFEB

Figure S1 related to Figure 1. C. elegans DKF-1/protein kinase D is necessary and sufficient for the activation of HLH-30. (A) qRT-PCR of *dkf-1 and dkf-2* in lysates from animals treated with empty vector control (EV) or *dkf-1* RNAi. Results are normalized to empty vector control animals. Data are mean ± SEM (two biological replicates, two technical replicates, n ≥ 3,000 per condition) * $p \leq 0.05$ (two-sample *t* test). **(B)** Lifespan of wild type and *hlh-30* mutant animals reared on *E. coli* HT115 carrying *dkf-1* RNAi or empty vector control prior to transfer to *E. coli* OP50. *** $p \leq 0.001$ (Log-Rank test). **(C)** Survival of wild type and *dkf-1*(ok2695) mutant animals infected with *S. aureus*. Shown is a representative experiment of three biological replicates. **(D)** qRT-PCR of *ilys-2* in wild type or in *pkc-1, pkc-2, tpa-1, or dkf-2* mutants. Results are normalized to wild type animals. Data are mean ± SEM (two biological replicates, two technical replicates, n ≥ 3,000 per condition). **(E)** Survival of animals reared on *E. coli* HT115 carrying *plc-1* RNAi or empty vector control (EV) RNAi. *** $p \leq 0.001$ (Log-Rank test). **(F)** Lifespan of animals fed *E. coli* carrying *plc-1* RNAi or empty vector control (EV) RNAi. *** $p \leq 0.001$ (Log-Rank test).

Figure S2 related to Figure 3. PKD1 and PKCα/γ are necessary for activation of TFEB by infection. TFEB-GFP RAW264.7 cells were preincubated with PKC and PKD inhibitors for 1 h previous to infection with *S. enterica* (MOI = 100) for 2 hours. Shown are representative images from one replicate, and quantification of three biological replicates of three technical replicates each. Scale bars = 100 µm. (**A**) DMSO control. (**A'**) detail. (**B**) *S. enterica* SL1344. (**B'**) detail. (**C**) 5 µM CRT0066101 (PKD inhibitor). (**C'**) detail. (**D**) 5 µM Bisindolylmaleimide IV (pan-PKC inhibitor). (**D'**) detail. (**E**) 10 µM PKCε Inhibitor Peptide (selectively inhibits PKCε). (**E'**) detail. (**F**) Percentage of cells with nuclear translocation was measured with Gen5 analysis software. (**G**) GFP intensity in nucleus compared to cytoplasm (N/C ratio) was measured using CellProfiler. Please see *Methods* for more detail. ** $p \leq 0.01$, *** $p \leq 0.001$ (One-way ANOVA followed by Tukey's post-hoc test).

Figure S3 related to Figure 4. PKC is necessary for TFEB activation after *S. aureus* infection. TFEB-FLAG RAW264.7 cells were incubated with inhibitor for 1 h prior to infection with *S. aureus* NCTC8325 (MOI = 10) for 2 h, followed by anti-FLAG immunofluorescence and Hoechst DNA staining. Shown are representative

images from one replicate, and quantification of three biological replicates of three technical replicates each. Scale bars = 100 µm. (**A, B**) DMSO control. (**C, D**) *S. aureus*. (**E, F**) *S. aureus* previously killed with 100 µg/ml gentamicin. (**G, H**) 5 µM Bisindolylmaleimide IV (pan-PKC inhibitor). (**I, J**) 5 µM Gö 6983 (pan-PKC inhibitor). (**K**) Percentage of cells with nuclear translocation was measured with Gen5 analysis software. (**L**) GFP intensity in nucleus compared to cytoplasm (N/C ratio) was measured using CellProfiler. Please see *Methods* for more detail. ** $p \leq 0.01$, *** $p \leq 0.001$ (One-way ANOVA followed by Tukey's post-hoc test).

Figure S4 related to Figure 5A, K. Whole immunoblot pictures with size markers for TFEB antibody. (**A, B**) RAW264.7 cells were infected with *S. enterica* SL1344 (MOI = 100) for 0 (control), 10, 20, 30, 60, and 120 min, lysed, and subjected to immunoblot analysis. (**A**) TFEB antibody. (**B**) TFEB antibody after SL1344 infection plus 10 µM kb-NB142-70 (specific PKD inhibitor). MW, molecular weight. The TFEB band is indicated on the right.