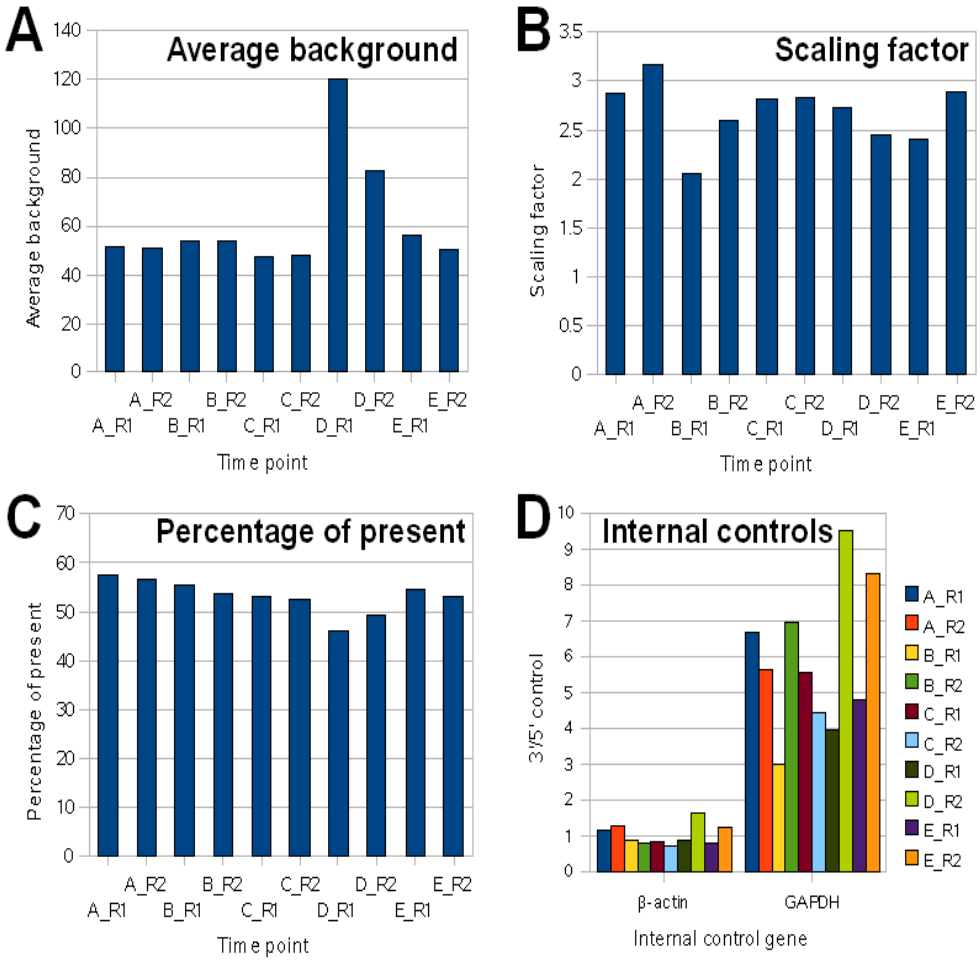
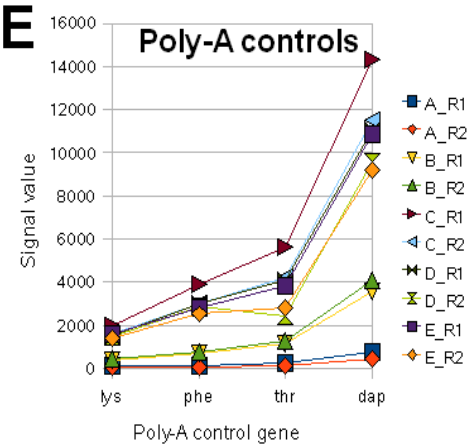
S1 Detail on quality assessment of microarray hybridization.

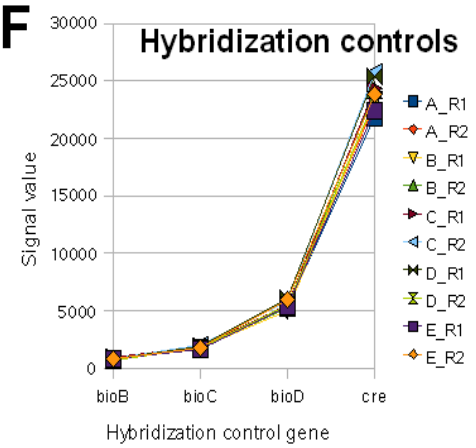
Background on quality parameters are provided in Affymetrix GeneChip Expression Analysis: Data Analysis Fundamentals (Page 36-40). (A) Average background, typical ranging from 20 to 100. (B) Scaling factor, usually around 3, less than 5 is considered acceptable; (C) Percent of probes detected; 50% is common. (D) Internal controls genes β-actin and GAPDH used to assess RNA sample and assay quality. Specifically, the ratio of the 3' probe set to the 5' probe set is generally no more than 3. However, a high 3' to 5' ratio of only one group of the internal control genes does not necessarily indicate RNA degradation. (E) Poly-A controls used to monitor the entire target labeling process. All controls should be called "Present" with increasing signal value in the order of lys, phe, thr, and dap. (F) Hybridization controls independent of RNA sample preparation, and used to evaluate sample hybridization; their signal values should reflect their relative concentrations (bioB:bioC:bioD:cre = 1.5:5:25:100). For (A) – (G), A-E at X-axis indicates five time point for collecting samples. R1 and R2 indicate biological replicate group 1 and 2, respectively. (G) Correlation efficiency between biological replicates at five time points for collecting samples. (H) Artifact bias detected by residual image. The value at a probe is calculated as: signal intensity after normalization – signal intensity prior to normalization. Red: positive residue. Blue: negative residue.


A
Average background
Average background
140
120
100
80
60
40
20
0
A_R2
B_R2
C_R2
D_R2
E_R2
A_R1
B_R1
C_R1
D_R1
E_R1
Time point
B
Scaling factor
Scaling factor
3.5
3
2.5
2
1.5
1
0.5
0
A_R2
B_R2
C_R2
D_R2
E_R2
A_R1
B_R1
C_R1
D_R1
E_R1
Time point
C
Percentage of present
Percentage of present
70
60
50
40
30
20
10
0
A_R2
B_R2
C_R2
D_R2
E_R2
A_R1
B_R1
C_R1
D_R1
E_R1
Time point
D
Internal controls
3'/5' control
10
9
8
7
6
5
4
3
2
1
0
β-actin
GAPDH
Internal control gene
A_R1
A_R2
B_R1
B_R2
C_R1
C_R2
D_R1
D_R2
E_R1
E_R2

E



F



G

Time point	A	B	C	D	E
Correlation efficiency between two biological replicates	0.987	0.990	0.979	0.940	0.974

H