Example 2 Suppose you are given the data below in a two by two table.

- > pm.ex<-pm(replicates, featureNames(replicates)[grep("205586_x_at", featureNames(replicates))])
- > pm.ex

REP1.CEL	REP2.CEL	REP3.CEL
134.3	110.3	138.0
265.0	304.5	290.3
254.3	277.8	285.3
73.5	86.3	73.8
145.0	157.5	179.3
202.0	209.0	207.5
228.3	237.3	224.5
319.3	343.3	405.8
829.5	736.5	820.0
102.5	118.3	122.8
138.3	96.3	103.0
	134.3 265.0 254.3 73.5 145.0 202.0 228.3 319.3 829.5 102.5	265.0 304.5 254.3 277.8 73.5 86.3 145.0 157.5 202.0 209.0 228.3 237.3 319.3 343.3 829.5 736.5 102.5 118.3

(0) iteration: Start with the raw data in a two-way table, I represents the row effect, J represent the column effect.

Ţ		J	
1	1	2	3
1	134.3	110.3	138.0
2	265.0	304.5	290.3
3	254.3	277.8	285.3
4	73.5	86.3	73.8
5	145.0	157.5	179.3
6	202.0	209.0	207.5
7	228.3	237.3	224.5
8	319.3	343.3	405.8
9	829.5	736.5	820.0
10	102.5	118.3	122.8
11	138.3	96.3	102.0

(1) iteration, step a: The previous row $a_i^{(0)}$, column $b_j^{(0)}$, and main effect $m^{(0)}$ are initialized to 0. Then, find the median of each row, $\Delta a_i^{(1)}$.

Kellie J. Archer, Ph.D.

BIOS 567 Statistical Methods for High-Throughput Genomic Data

I		J		Row median $\Delta a_{i}^{(1)}$	Previous row effect $a_i^{(0)}$
	1	2	3		
1	134.3	110.3	138.0	134.3	0
2	265.0	304.5	290.3	<mark>290.3</mark>	0
3	254.3	277.8	285.3	<mark>277.8</mark>	0
4	73.5	86.3	73.8	<mark>73.8</mark>	0
5	145.0	157.5	179.3	157.5	0
6	202.0	209.0	207.5	207.5	0
7	228.3	237.3	224.5	228.3	0
8	319.3	343.3	405.8	343.3	0
9	829.5	736.5	820.0	820.0	0
10	102.5	118.3	122.8	118.3	0
11	138.3	96.3	103.0	103.0	0
Prev	0	0	0		$m^{(0)} = 0$
Column					III V
Effect					
$b_j^{(0)}$					

(1) **iteration, step b:** Row polish by subtracting the row median values from the corresponding row observations. Find the column medians after the row polish, $\Delta b_i^{(1)}$.

				Row median	Previous row effect
I		J		$\Delta a_{i}^{(1)}$	$a_i^{(0)}$
	1	2	3		
1	0	-24.0	3.7	134.3	0
2	-25.3	14.2	0	290.3	0
3	-23.5	0	7.5	277.8	0
4	-0.3	12.5	0	73.8	0
5	-12.5	0	21.8	157.5	0
6	-5.5	1.5	0	207.5	0
7	0	9.0	-3.8	228.3	0
8	-24.0	0	62.5	343.3	0
9	9.5	-83.5	0	820.0	0
10	-15.8	0	4.5	118.3	0
11	35.3	-6.7	0	103.0	0
Column median $\Delta b_j^{(1)}$	<mark>-5.5</mark>	0	0		
Prev Column Effect	0	0	0	0	$m^{(0)}=0$
$b_{i}^{(0)}$					

(1) iteration, step c: Column polish by subtracting the column median values from the corresponding column observations.

(1) herahon, step c. (zorumni po	ilish by su	on acting t	ne column median	values from the com-
		-		Row median	Previous row effect
I	J			$\Delta a_{i}^{(1)}$	$a_i^{(0)}$
	1	2	3		
1	5.5	-24.0	3.7	134.3	0
2	-19.8	14.2	0.0	290.3	0
3	-18.0	0.0	7.5	277.8	0
4	5.2	12.5	0.0	73.8	0
5	-7.0	0.0	21.8	157.5	0
6	0.0	1.5	0.0	207.5	0
7	5.5	9.0	-3.8	228.3	0
8	-18.5	0.0	62.5	343.3	0
9	15.0	-83.5	0.0	820.0	0
10	-10.3	0.0	4.5	118.3	0
11	40.8	-6.7	0.0	102.0	0
Column median $\Delta b_j^{(1)}$	-5.5	0	0		
Prev Column Effect	0	0	0		$m^{(0)}=0$
$b_{j}^{(0)}$					

(1) iteration, step d: Estimate the effects by

$$\begin{split} \Delta m_a^{(1)} = & \text{median}(a_i^{(0)} + \Delta a_i^{(1)}) = 207.5 \\ \Delta m_b^{(1)} = & \text{median}(b_j^{(0)}) = 0 \\ m^{(1)} = & m^{(0)} + \Delta m_a^{(1)} + \Delta m_b^{(1)} = 0 + 207.5 + 0 = 207.5 \\ a_i^{(1)} = & a_i^{(0)} + \Delta a_i^{(1)} - \Delta m_a^{(1)} \\ b_j^{(1)} = & b_j^{(0)} + \Delta b_j^{(1)} - \Delta m_b^{(1)} \end{split} \qquad \qquad \text{(column effect estimates after 1st iteration)} \end{split}$$

I	J		Row median $\Delta a_{\rm i}^{(1)}$	Previous row effect $a_i^{(0)}$	$a_{i}^{(0)} + \Delta a_{i}^{(1)}$	$a_i^{(1)} = a_i^{(0)} + \Delta a_i^{(1)} - \Delta m_a^{(1)}$	
	1	2	3				
1	5.5	-24.0	3.7	134.3	0	134.3	<mark>-73.2</mark>
2	-19.8	14.2	0.0	290.3	0	290.3	82.8
3	-18.0	0.0	7.5	277.8	0	277.8	<mark>70.3</mark>
4	5.2	12.5	0.0	73.8	0	73.8	-133.7
5	-7.0	0.0	21.8	157.5	0	157.5	<mark>-50.0</mark>
6	0.0	1.5	0.0	207.5	0	207.5	0.0
7	5.5	9.0	-3.8	228.3	0	228.3	20.8
8	-18.5	0.0	62.5	343.3	0	343.3	135.8
9	15.0	-83.5	0.0	820.0	0	820.0	612.5
10	-10.3	0.0	4.5	118.3	0	118.3	<mark>-89.2</mark>
11	40.8	-6.7	0.0	102.0	0	102.0	-104.5
Column median $\Delta b_{j}^{(1)}$	-5.5	0	0				
Prev Column Effect $b_{j}^{(0)}$	0	0	0		$m^{(0)}=0$		
$b_j^{(1)} = b_j^{(0)} + \Delta b_j^{(1)} - \Delta m_b^{(1)}$	<mark>-5.5</mark>	0	0				

(2) **iteration, step a:** For the second iteration, retain the previous row $a_i^{(1)}$, column $b_j^{(1)}$, and main effect $m^{(1)}$ as estimates of row and column and main effects. Then, find the median of each row.

and main effects. Then, find the median of each row.							
		_		Row median	Previous row effect		
I		J		$\Delta a_{i}^{(2)}$	$\mathbf{a}_{\mathrm{i}}^{(1)}$		
	1	2	3				
1	5.5	-24.0	3.7	3.7	-73.2		
2	-19.8	14.2	0.0	0.0	82.8		
3	-18.0	0.0	7.5	0.0	70.3		
4	5.2	12.5	0.0	5.2	-133.7		
5	-7.0	0.0	21.8	0.0	-50.0		
6	0.0	1.5	0.0	0.0	0.0		
7	5.5	9.0	-3.8	<mark>5.5</mark>	20.8		
8	-18.5	0.0	62.5	0.0	135.8		
9	15.0	-83.5	0.0	0.0	612.5		
10	-10.3	0.0	4.5	0.0	-89.2		
11	40.8	-6.7	0.0	0.0	-104.5		
Prev Column Effect	-5.5	0	0		$m^{(1)}=207.5$		
$b_j^{(1)}$							

(2) **iteration**, **step b:** Row polish by subtracting the row median values from the corresponding row observations. Find the column medians after the row polish.

after the row polish.				Row median	Previous row effect
		J			
I		,		$\Delta a_{i}^{(2)}$	$\mathbf{a}_{\mathrm{i}}^{(1)}$
	1	2	3		
1	1.8	-27.7	0.0	3.7	-73.2
2	-19.8	14.2	0.0	0.0	82.8
3	-18.0	0.0	7.5	0.0	70.3
4	0.0	7.3	-5.2	5.2	-133.7
5	-7.0	0.0	21.8	0.0	-50.0
6	0.0	1.5	0.0	0.0	0.0
7	0.0	3.5	-9.3	5.5	20.8
8	-18.5	0.0	62.5	0.0	135.8
9	15.0	-83.5	0.0	0.0	612.5
10	-10.3	0.0	4.5	0.0	-89.2
11	40.8	-6.7	0.0	0.0	-104.5
Column median $\Delta b_j^{(2)}$	0	0	0		
Prev Column Effect	-5.5	0	0	0	$m^{(1)}=207.5$
$b_j^{(1)}$					111 207.5

⁽²⁾ iteration, step c: Column polish by subtracting the column median values from the corresponding column observations.

(2) iteration, step d: Estimate the effects by

$$\begin{split} \Delta m_a^{(2)} &= median(a_i^{(1)} + \Delta a_i^{(2)}) = 0 \\ \Delta m_b^{(2)} &= median(b_j^{(1)}) = 0 \\ m^{(2)} &= m^{(1)} + \Delta m_a^{(2)} + \Delta m_b^{(2)} = 207.5 + 0 + 0 = 207.5 \\ a_i^{(2)} &= a_i^{(1)} + \Delta a_i^{(2)} - \Delta m_a^{(2)} \\ b_j^{(2)} &= b_j^{(1)} + \Delta b_j^{(2)} - \Delta m_b^{(2)} \end{split} \qquad \qquad \text{(row effect estimates after 1st iteration)} \end{split}$$

I	J		Row median $\Delta a_i^{(2)}$	Previous row effect $a_i^{(1)}$	$a_{i}^{(1)} + \Delta a_{i}^{(2)}$	$a_i^{(2)} = a_i^{(1)} + \Delta a_i^{(2)} - \Delta m_a^{(2)}$	
	1	2	3				
1	1.8	-27.7	0.0	3.7	-73.2	-69.5	<mark>-69.5</mark>
2	-19.8	14.2	0.0	0.0	82.8	82.8	82.8
3	-18.0	0.0	7.5	0.0	70.3	70.3	<mark>70.3</mark>
4	0.0	7.3	-5.2	5.2	-133.7	-128.5	<mark>-128.5</mark>
5	-7.0	0.0	21.8	0.0	-50.0	-50.0	<mark>-50.0</mark>
6	0.0	1.5	0.0	0.0	0.0	0.0	<mark>0.0</mark>
7	0.0	3.5	-9.3	5.5	20.8	26.3	<mark>26.3</mark>
8	-18.5	0.0	62.5	0.0	135.8	135.8	135.8
9	15.0	-83.5	0.0	0.0	612.5	612.5	<mark>612.5</mark>
10	-10.3	0.0	4.5	0.0	-89.2	-89.2	-89.2
11	40.8	-6.7	0.0	0.0	-104.5	-104.5	<mark>-104.5</mark>
Column median $\Delta b_{j}^{(2)}$	0	0	0				
Prev Column Effect $b_j^{(1)}$	-5.5	0	0		m ⁽¹⁾ =207.5		
$b_j^{(2)} = b_j^{(1)} + \Delta b_j^{(2)} - \Delta m_b^{(2)}$	-5.5	0	0				

Note that summing the main effect, row effect, column effect, and residual yields the original observation $y_{11,2} = 207.5 - 104.5 + 0 - 6.7 = 96.3$. The intensity for chips 1, 2, and 3 would be 207.5 - 5.5 = 202; 207.5 - 0 = 207.5; and 207.5 - 0 = 207.5 respectively. The log2 intensities would be $\log 2(202) = 7.658212$ for chip 1 and $\log (207.5) = 7.696968$ for chips 2 and 3.