Oligosaccharide Analysis – Arm Comparison

Methods

Due to the presence of observations below the limit of detection (LOD), analyses comparing oligosaccharide levels between arms and days were conducted using lognormal tobit models (Tobin, 1958), in which below-LOD observations were treated as censored at the lowest observed value for that oligosaccharide [i.e. for the purposes of modelling we're saying all we know about that data point is that it's lower than the lowest observed value].

Batch effects were assessed by comparing observations at day 0 between arms T1 and T2 using lognormal tobit models, including covariates for arm and subject. As most oligosaccharides differed significantly between these arms at day 0, batch effects were assumed to be non-negligible and data from arm T1/batch 1 were excluded from subsequent analyses.

Differences between arms in changes over time in oligosaccharide profiles were modeled using lognormal tobit models, including covariates for day, arm, the interaction between day and arm, and subject.

Analyses were conducted using R, version 3.4.0.

Results

Results of lognormal tobit models comparing changes over time between arms are in .csv files where the file name is the oligosaccharide name. These files include the following columns:

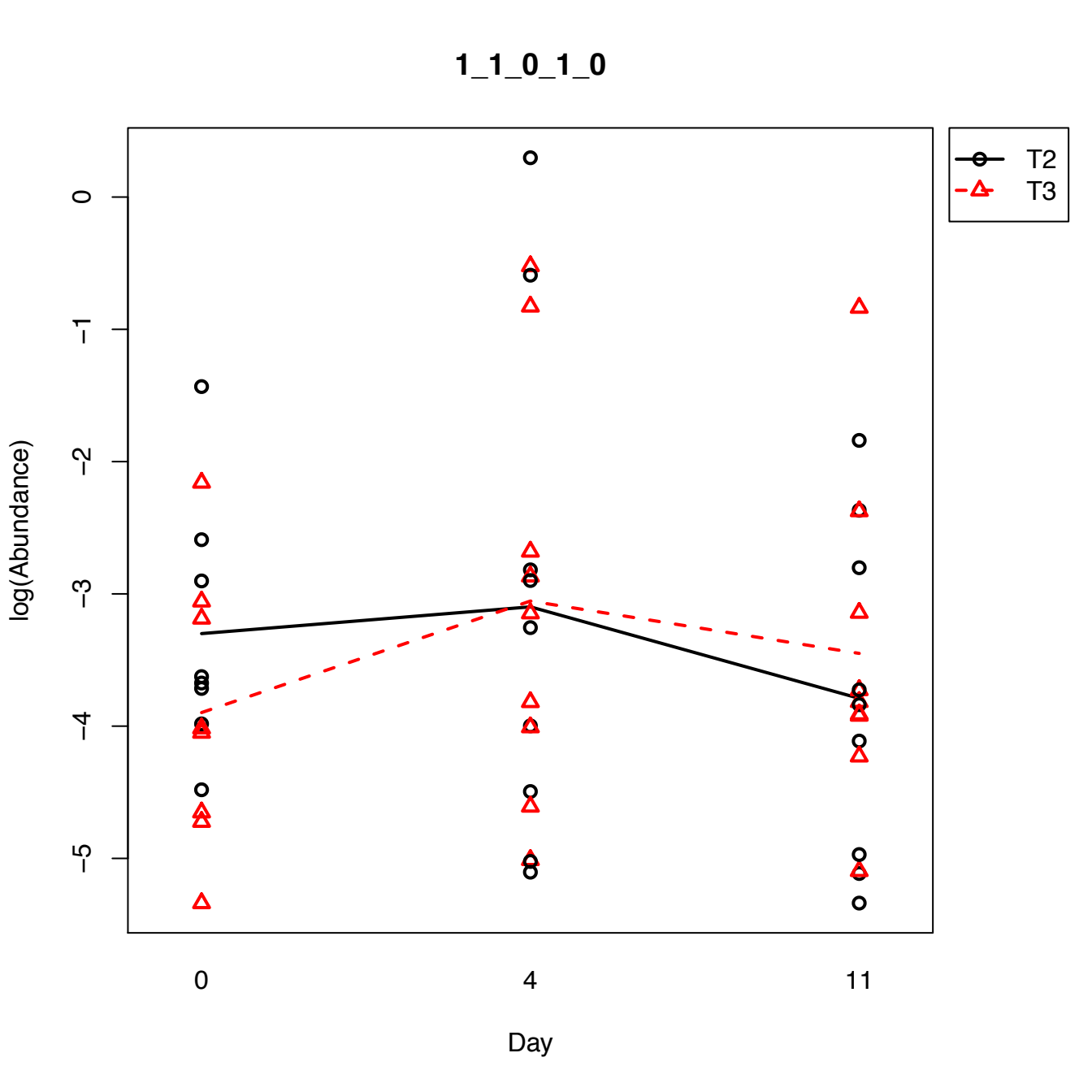
- Contrast
- GMR (95% CI): The geometric mean ratio (GMR) is the exponentiated regression coefficient from the lognormal tobit model. A GMR of 1.1, for example, would mean that abundance of that oligosaccharide is 10% higher in a given group compared to the reference group. The GMR is reported along with its 95% confidence interval
- PValue: P-value (unadjusted) for the indicated contrast from the lognormal tobit model

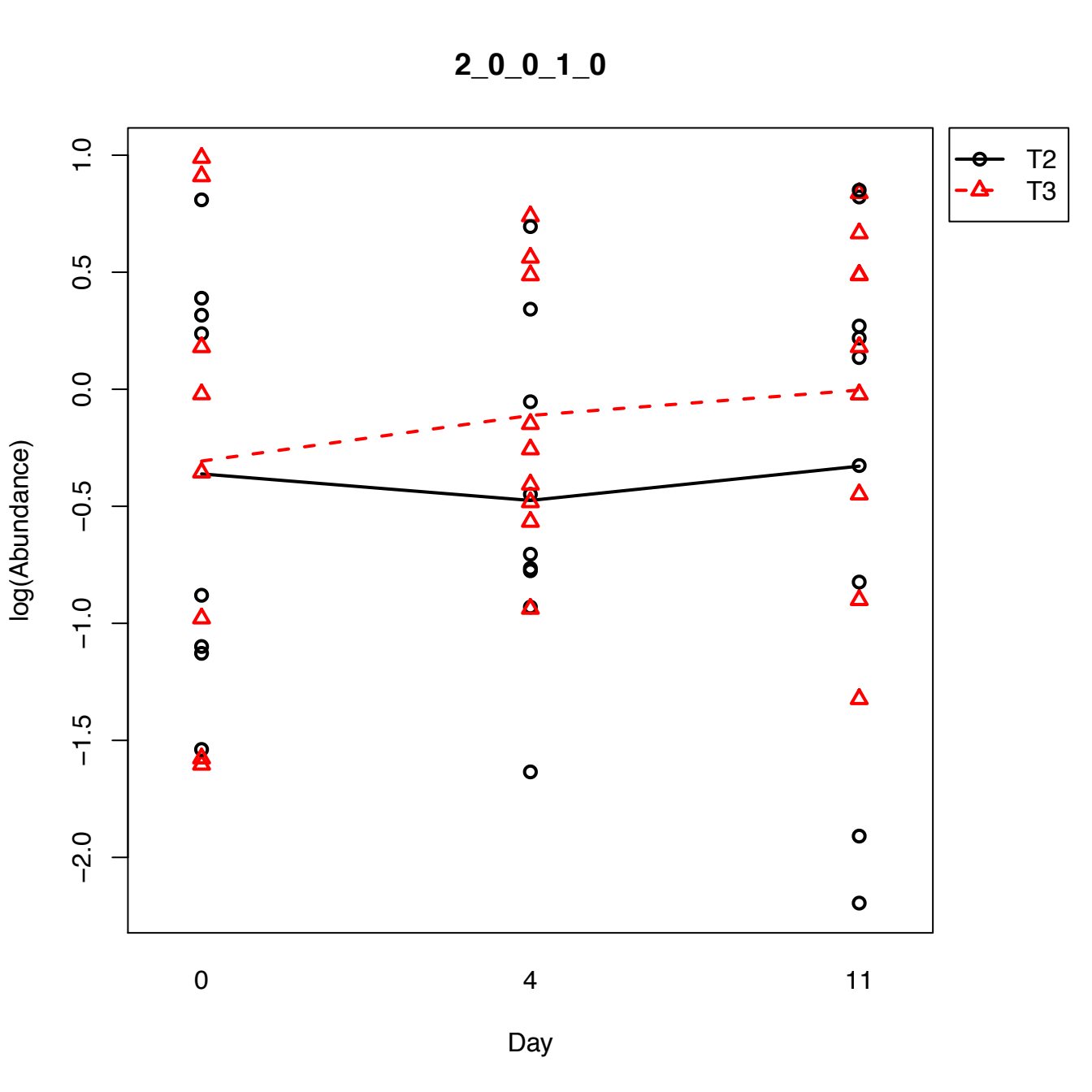
Plots of abundance over time by arm for each oligosaccharide are in the file "Abundance_by_Arm_and_Time.pdf":

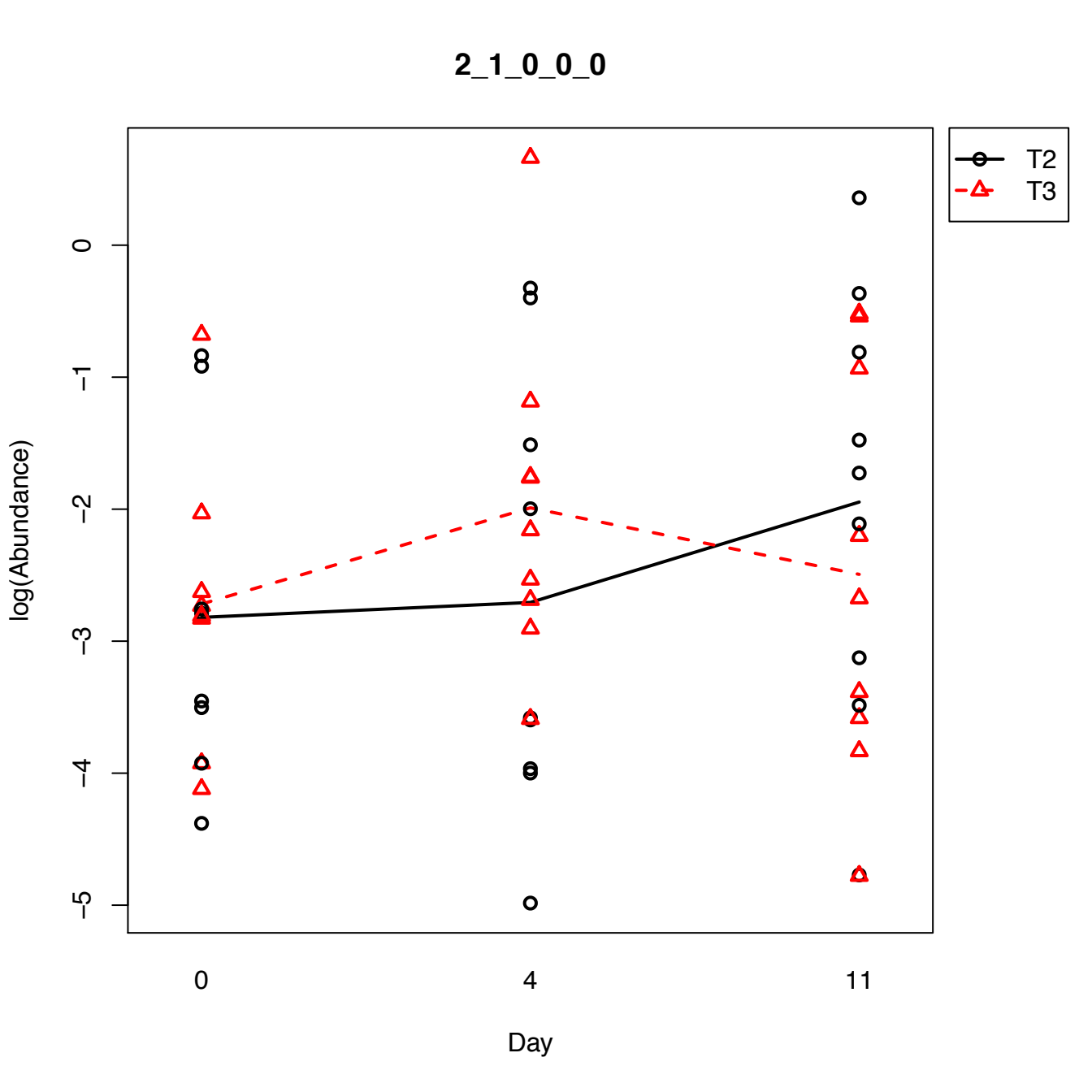
Black circles show the log(abundance) for each subject on arm T2 and red triangles show the log abundance for each subject on arm T3. The solid black line shows the mean log(abundance) for arm T2 and the dashed red line shows the mean log(abundance) for arm T3. For the purposes of plotting below-LOD observations were imputed at the lowest observed value.

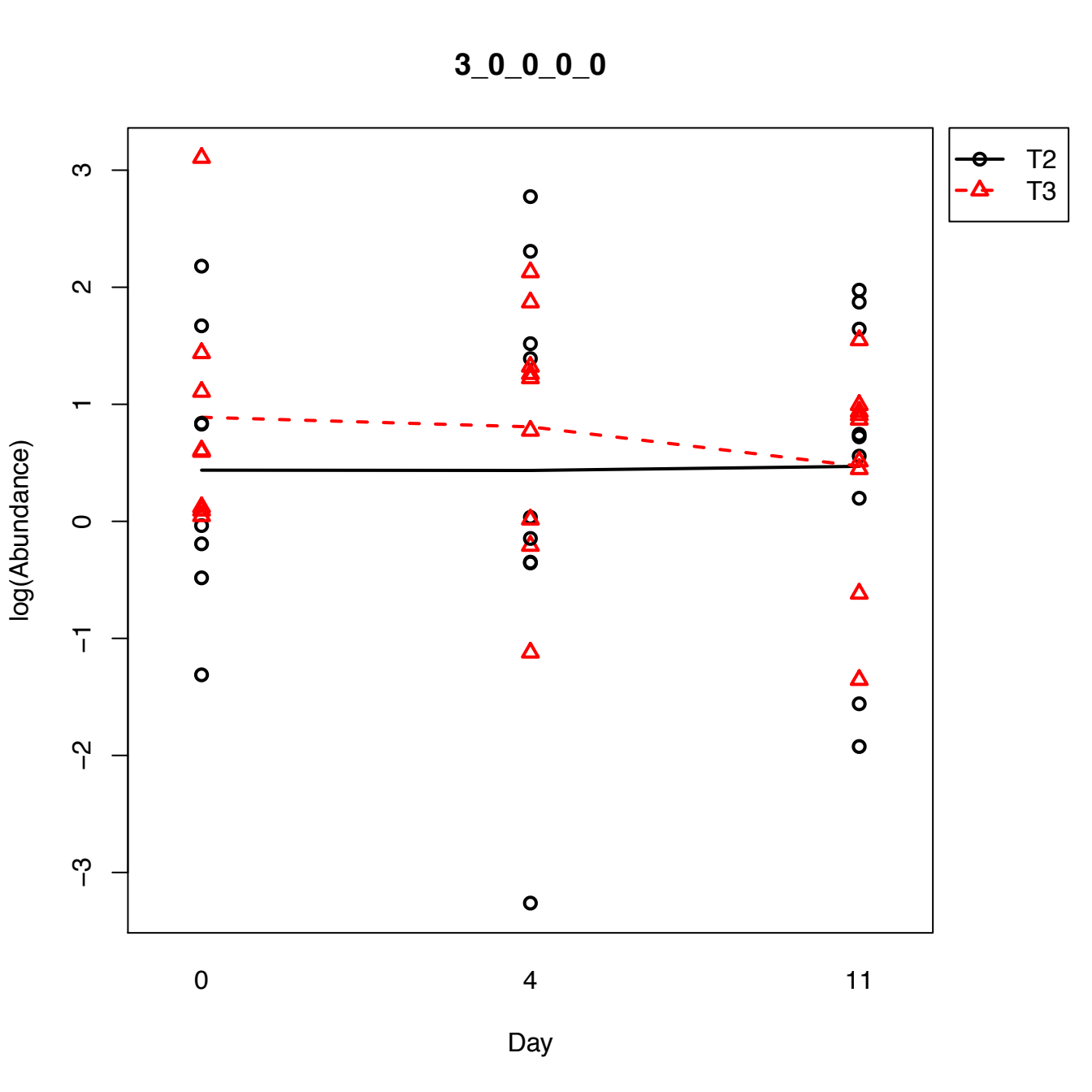
References

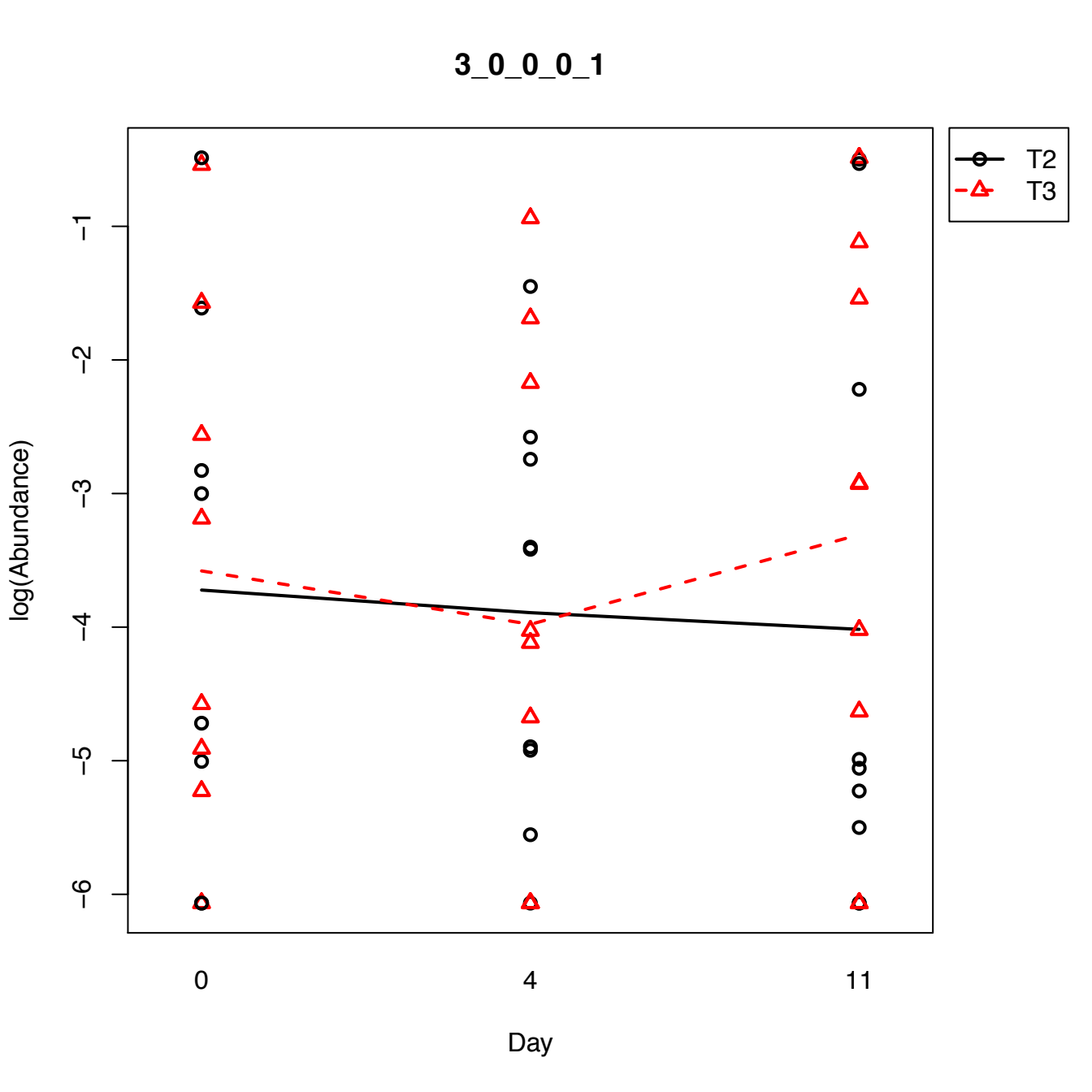
Tobin, James (1958). "Estimation of relationships for limited dependent variables". *Econometrica*. **26** (1): 24–36.

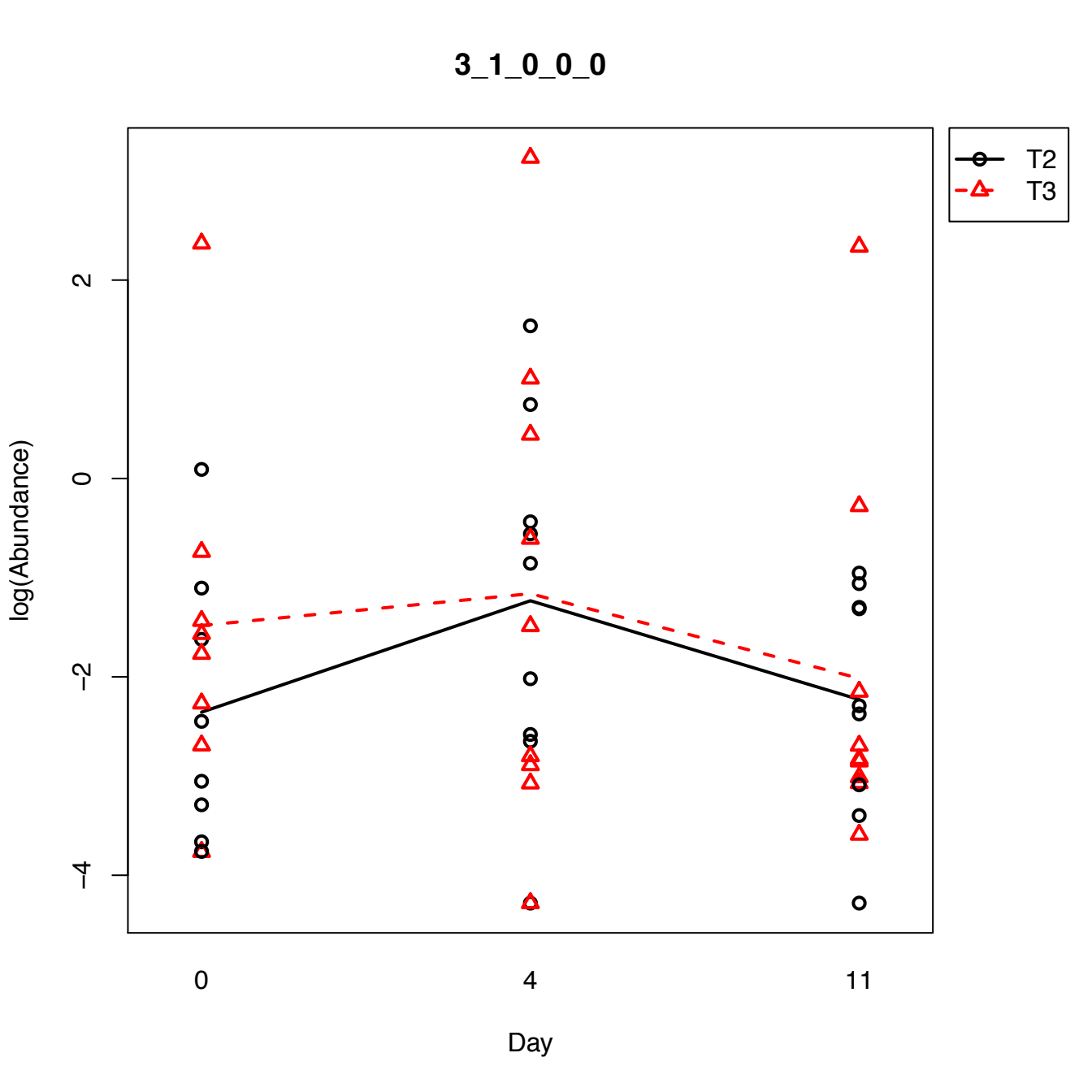

1_1_0_1_0
T2
T3
log(Abundance)
0
−1
−2
−3
−4
−5
0
4
11
Day

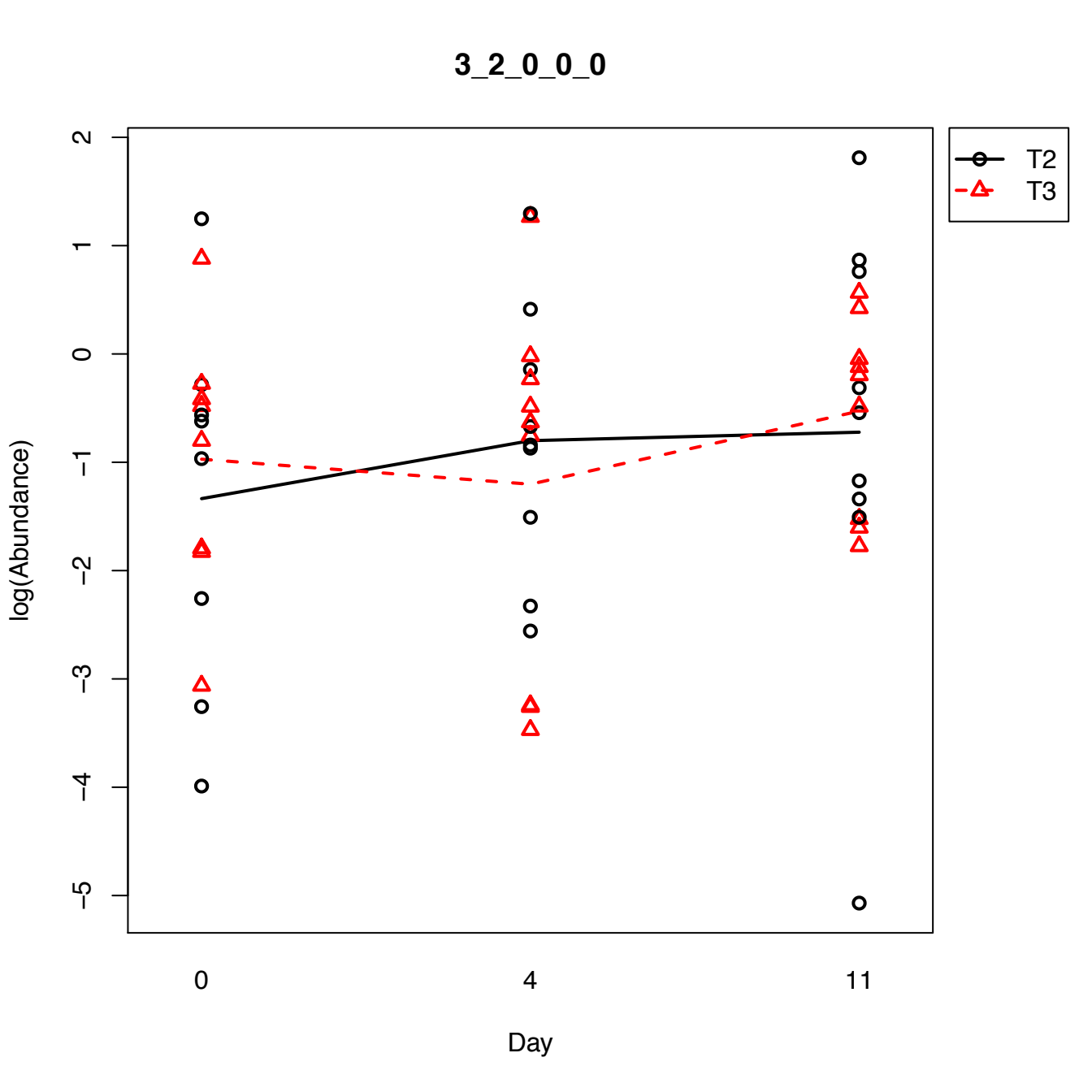

2_0_0_1_0
log(Abundance)
1.0
0.5
0.0
−0.5
−1.0
−1.5
−2.0
0
4
11
Day
T2
T3

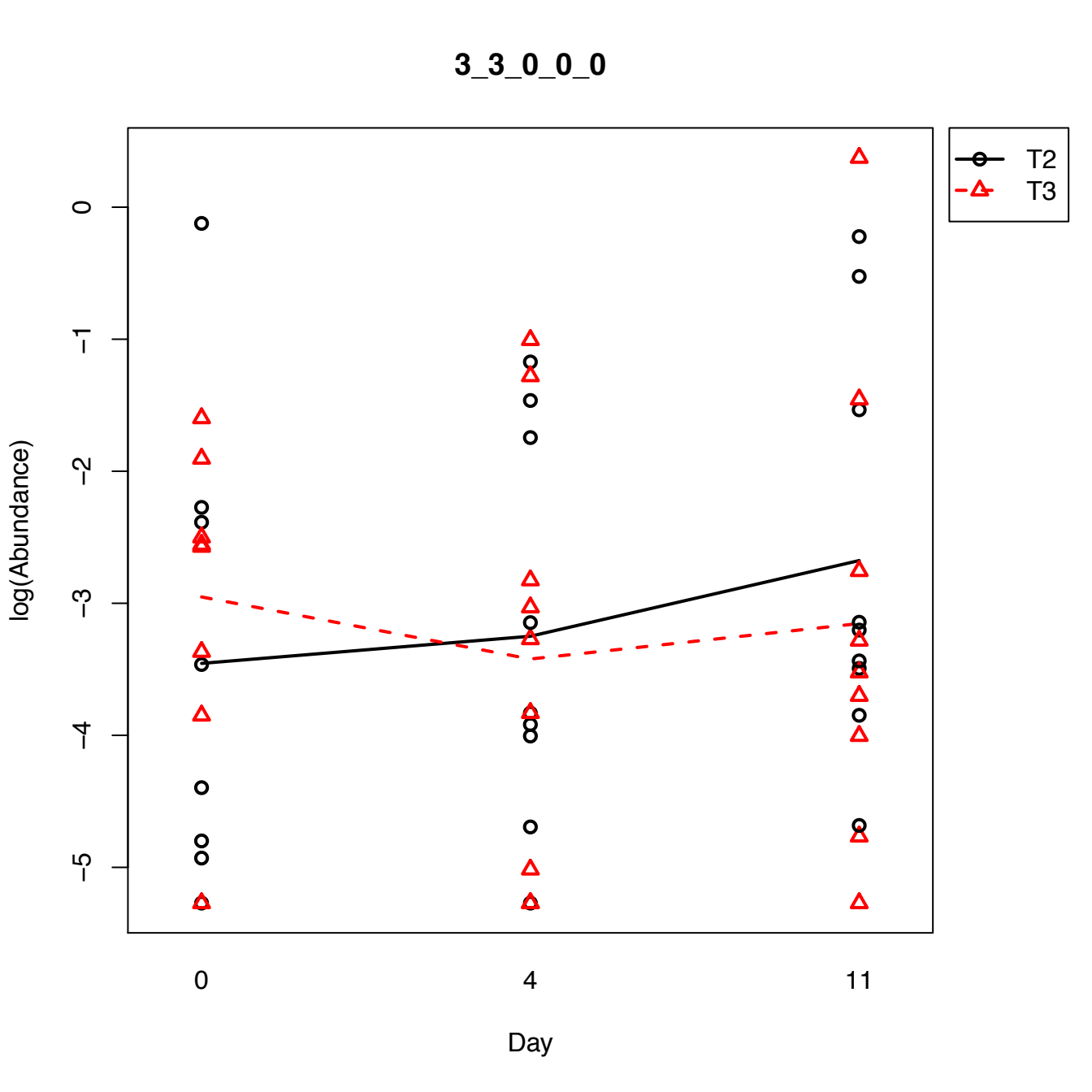

2_1_0_0_0
T2
T3
log(Abundance)
0
−1
−2
−3
−4
−5
0
4
11
Day

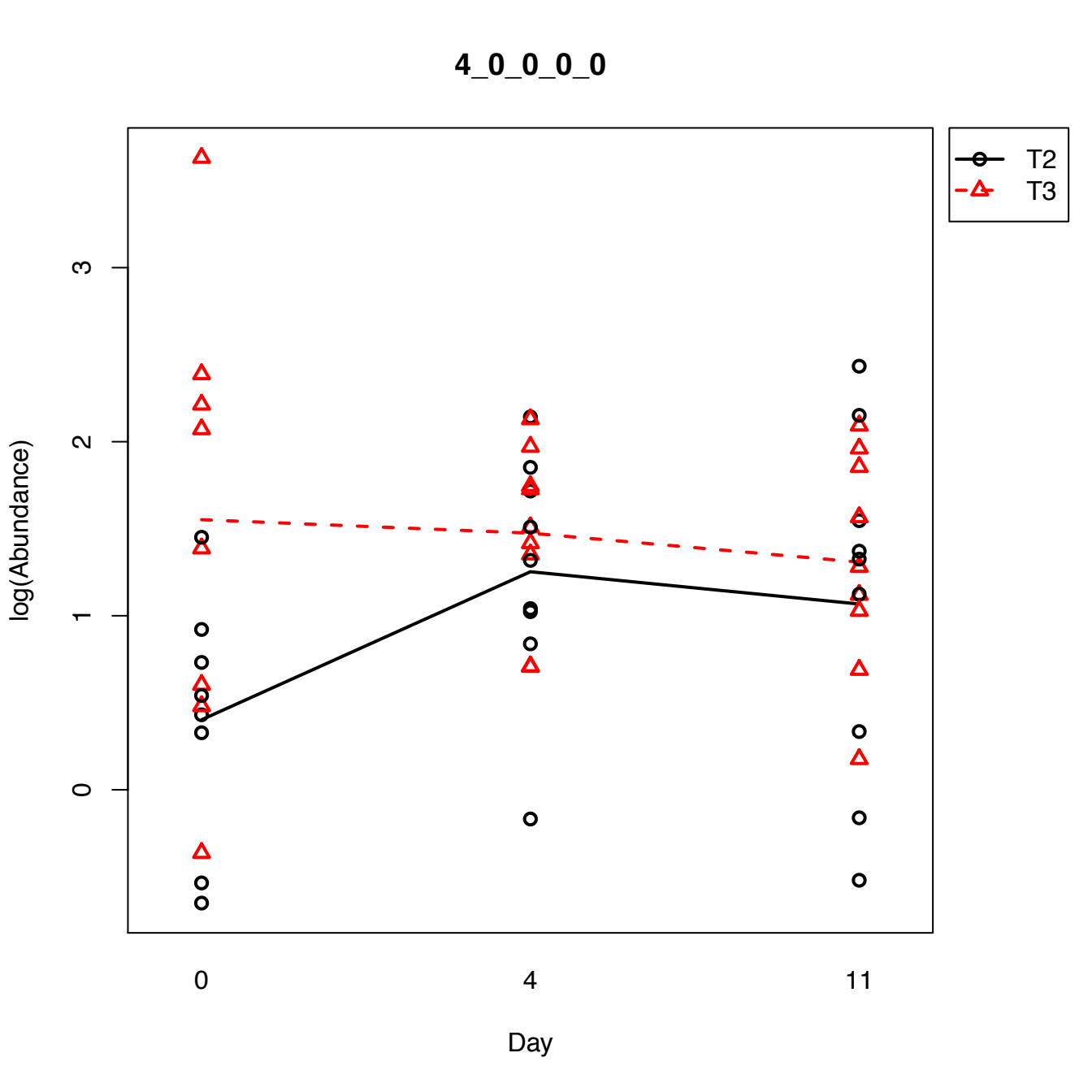

3_0_0_0_0
log(Abundance)
3
2
1
0
−1
−2
−3
0
4
11
Day
T2
T3

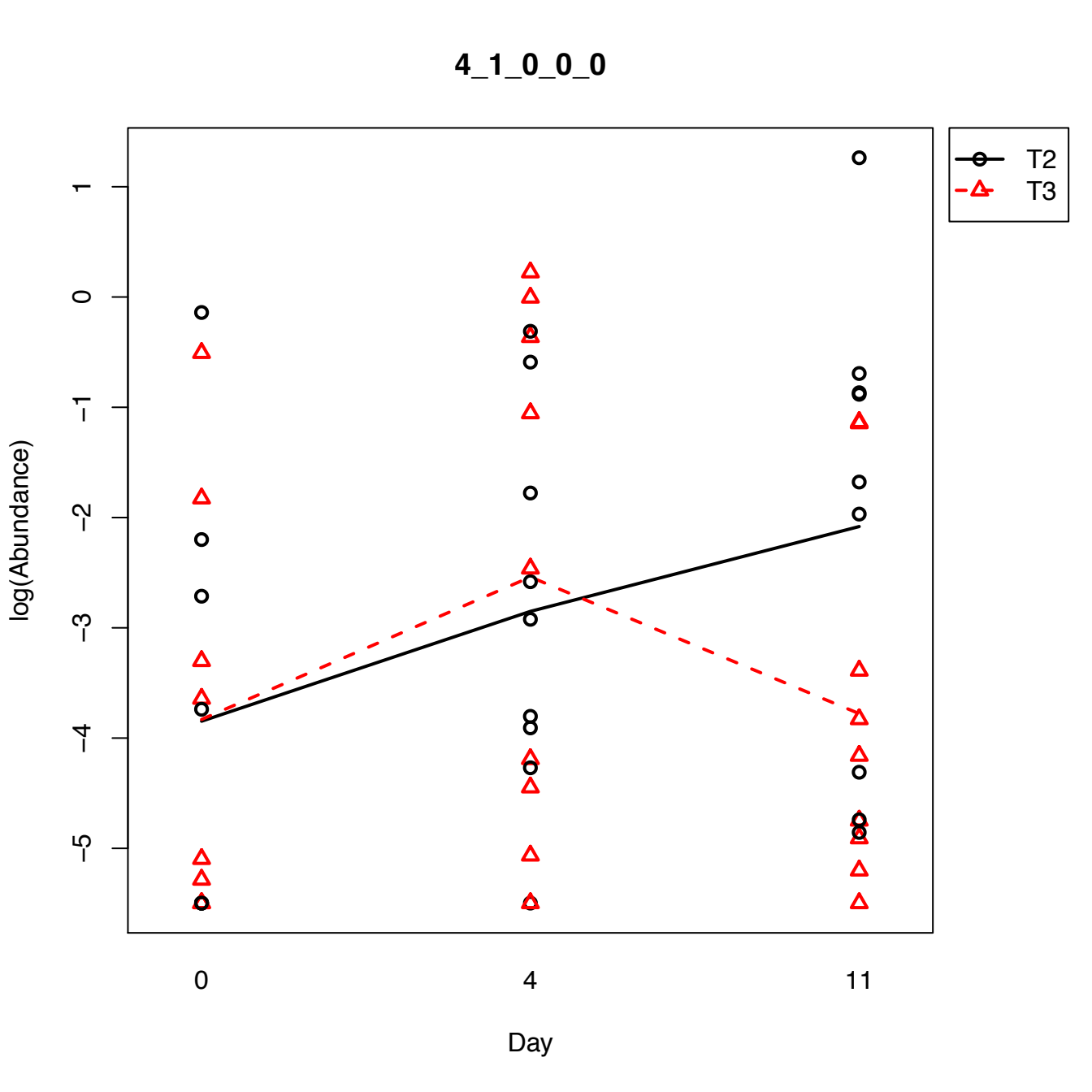

3_0_0_0_1
T2
T3
log(Abundance)
−1
−2
−3
−4
−5
−6
0
4
11
Day

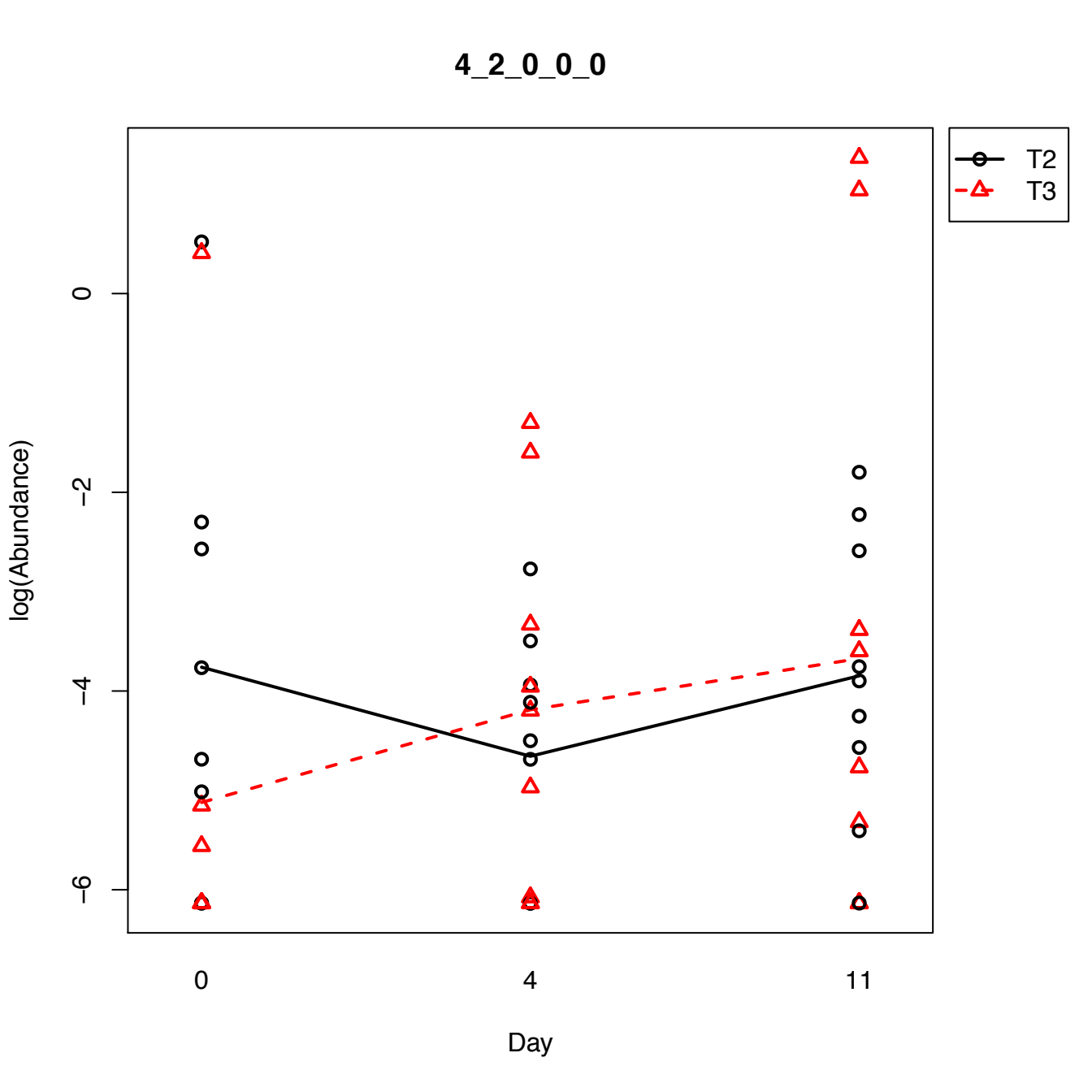

3_1_0_0_0
T2
T3
log(Abundance)
2
0
−2
−4
0
4
11
Day

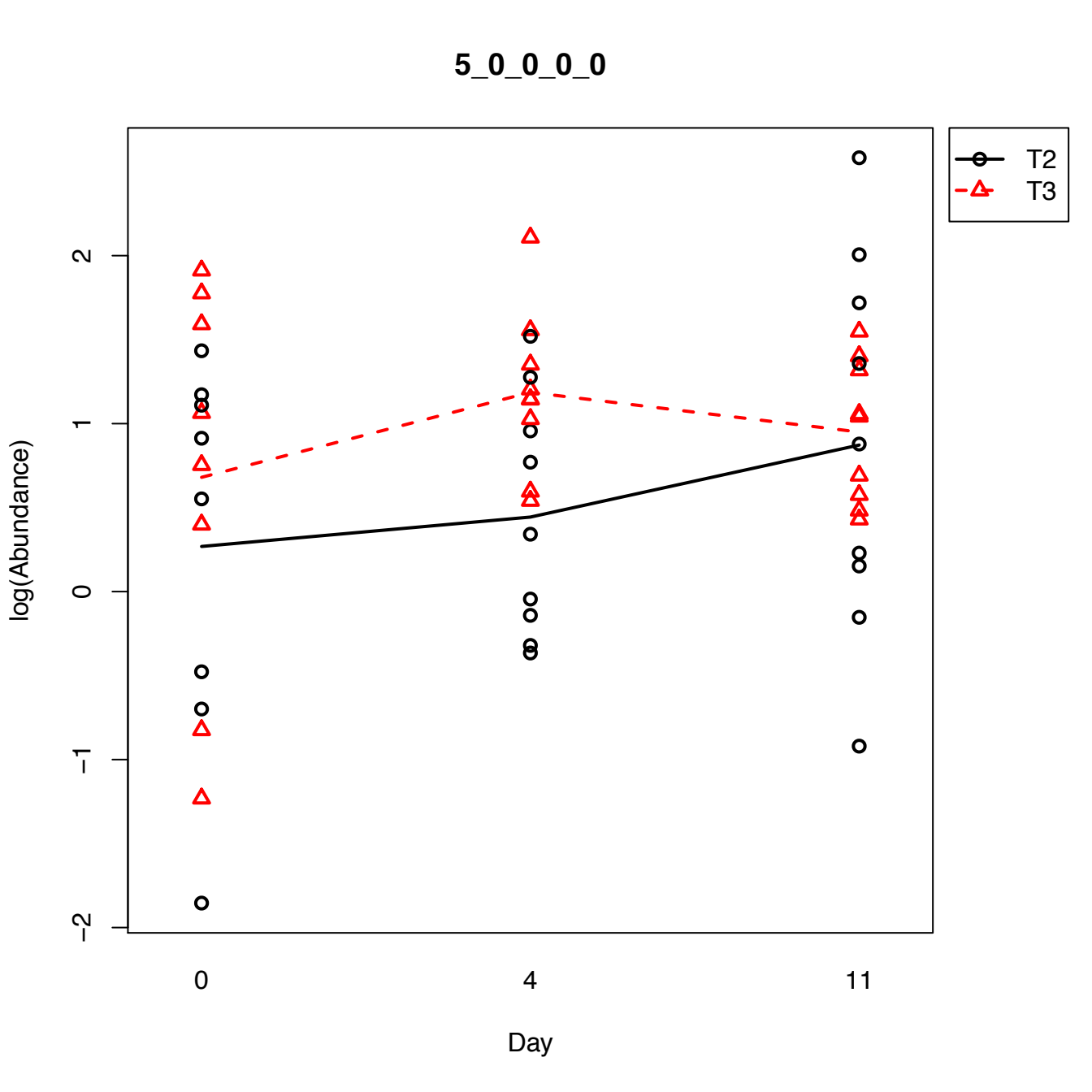

3_2_0_0_0
log(Abundance)
2
1
0
−1
−2
−3
−4
−5
0
4
11
Day
T2
T3

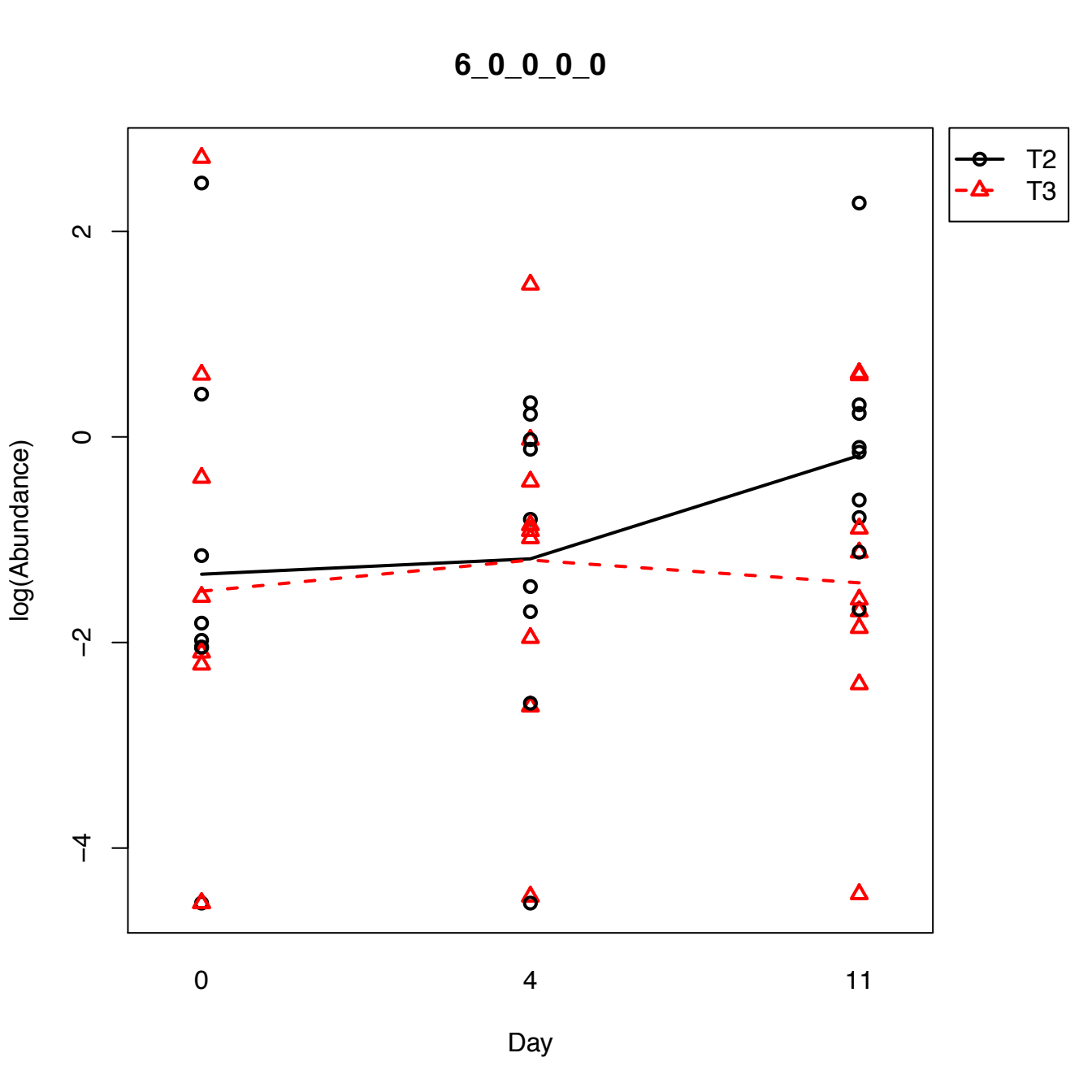

3_3_0_0_0
T2
T3
log(Abundance)
0
−1
−2
−3
−4
−5
0
4
11
Day


4_0_0_0_0
T2
T3
log(Abundance)
0
1
2
3
0
4
11
Day


4_1_0_0_0
T2
T3
log(Abundance)
1
0
−1
−2
−3
−4
−5
0
4
11
Day


4_2_0_0_0
T2
T3
log(Abundance)
0
−2
−4
−6
0
4
11
Day


5_0_0_0_0
T2
T3
log(Abundance)
2
1
0
−1
−2
0
4
11
Day


6_0_0_0_0
T2
T3
log(Abundance)
2
0
−2
−4
0
4
11
Day

Oligosaccharide Analysis – Enzyme Correlation

Methods

The correlations of each oligosaccharide with each enzyme were modeled using lognormal tobit models, with oligosaccharide level as the response and log(enzyme level) and subject as covariates.

Analyses were conducted using R, version 3.4.0.

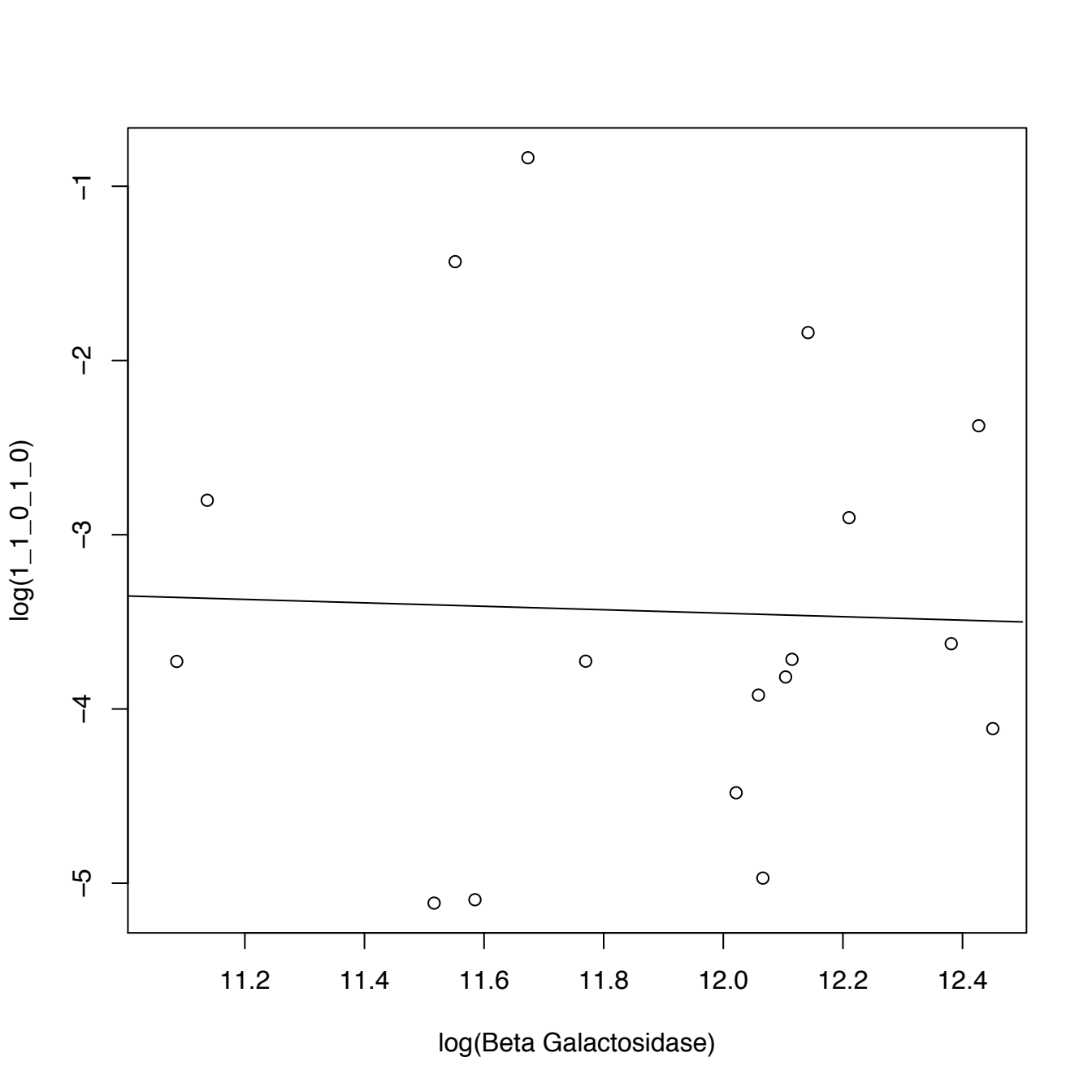
Results

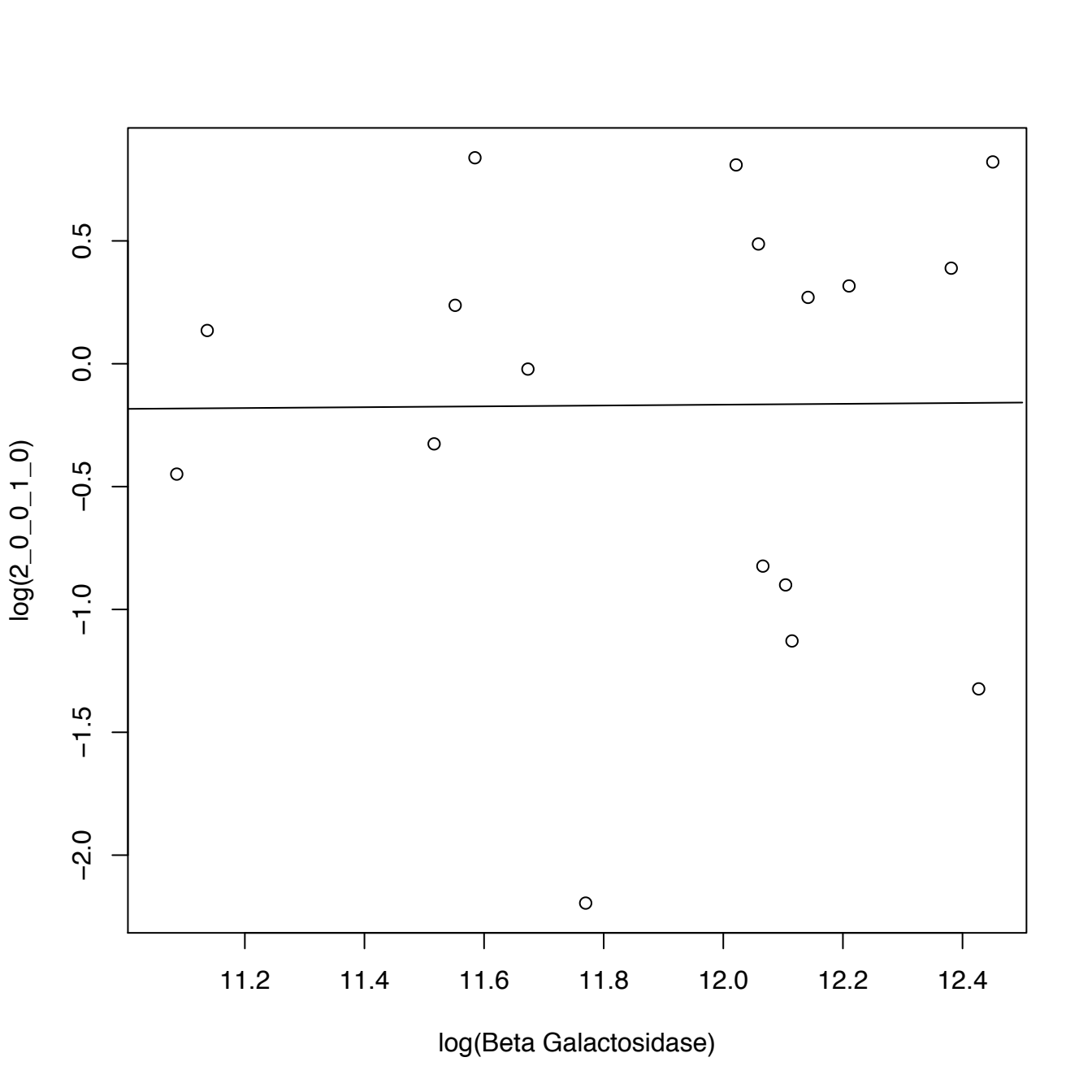
Results of the above lognormal tobit models are in the file "Oligosaccharide_by_enzyme_regression.csv". This file includes the following columns:

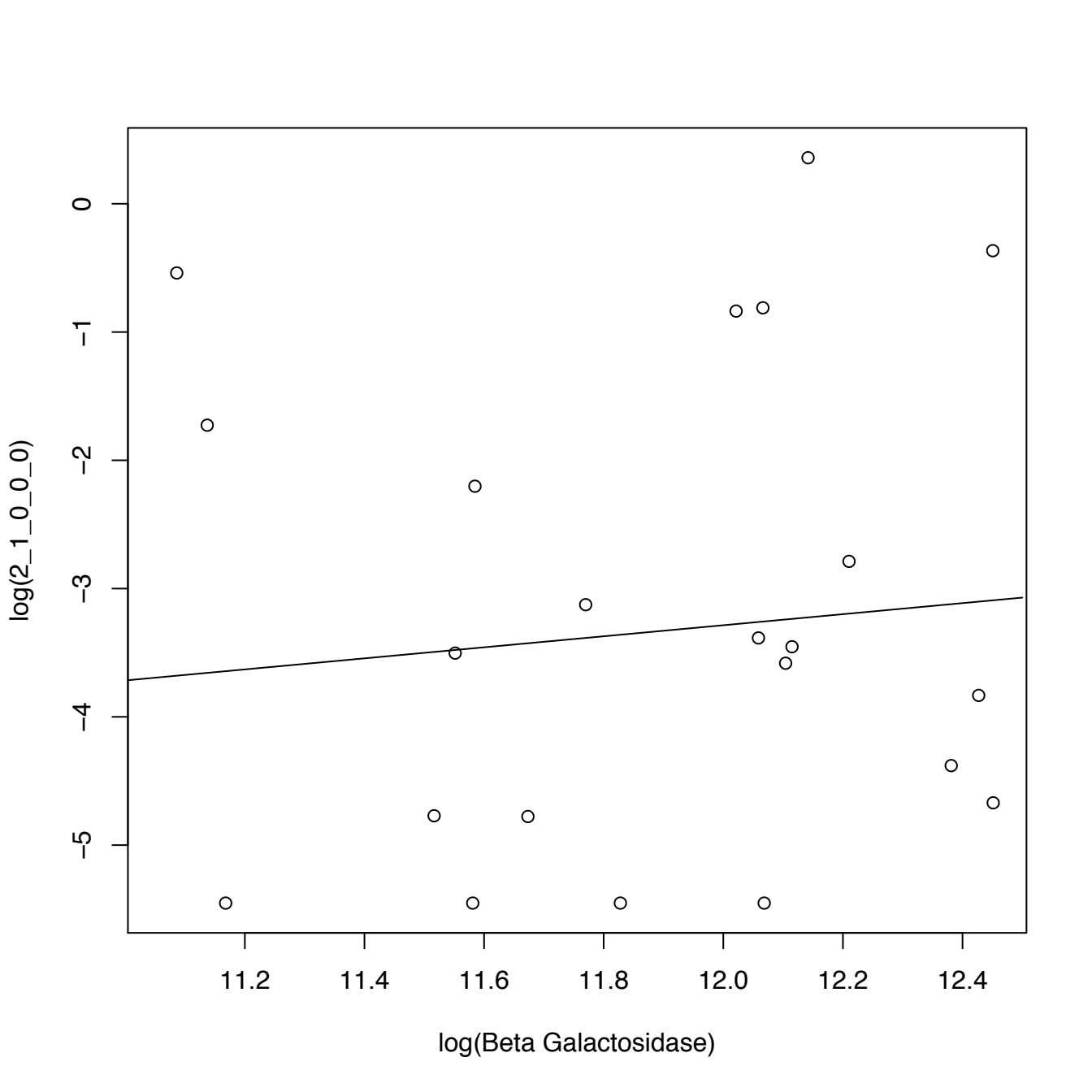
- Oligosaccharide
- Enzyme
- GMR (95% CI): The geometric mean ratio (GMR) is the exponentiated regression coefficient from the lognormal tobit model. The GMR is reported along with its 95% confidence interval
- PValue: P-value (unadjusted) for the indicated contrast from the lognormal tobit model

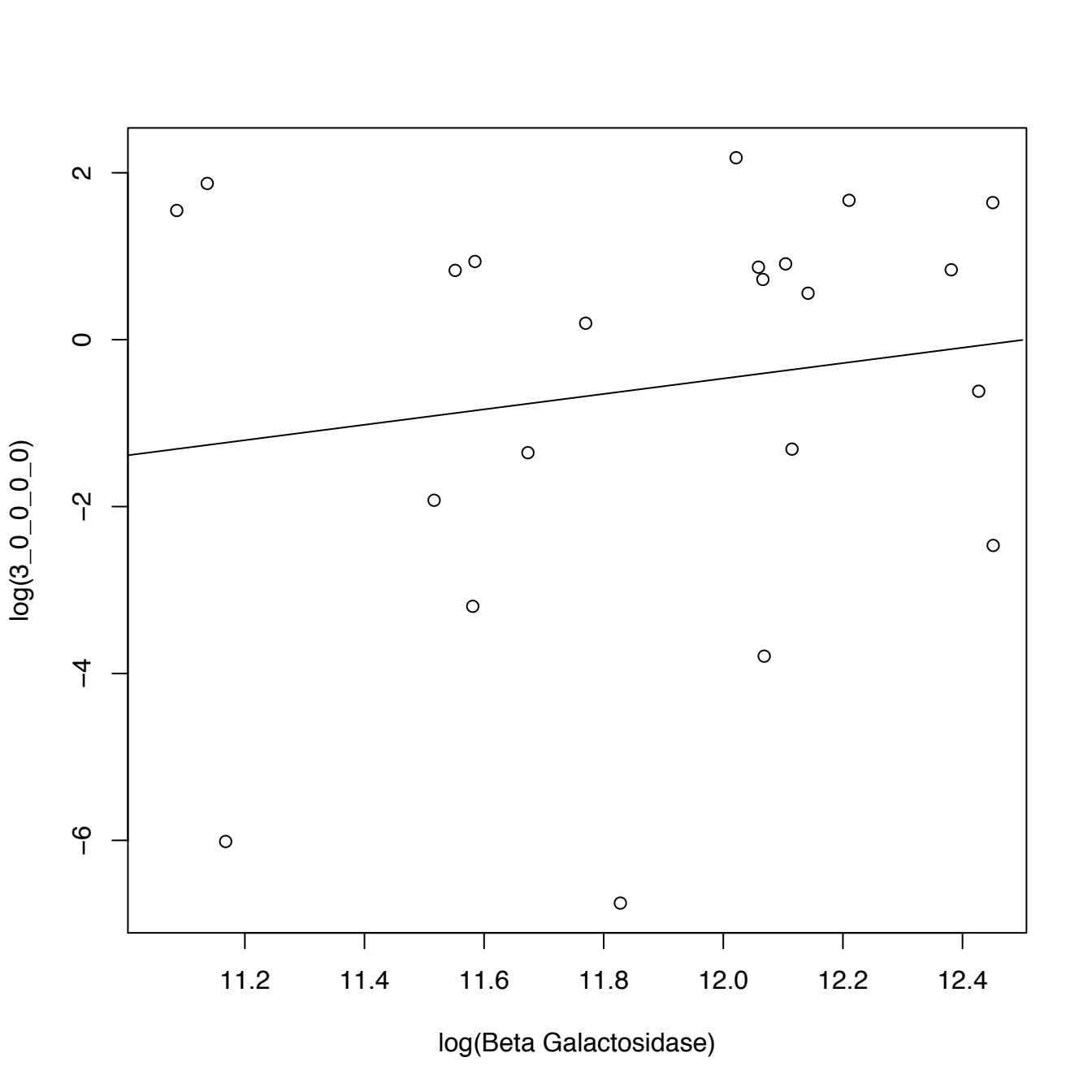
Plots of oligosaccharide levels by enzyme levels are in the files "Oligosaccharide Abundance_by_Beta_galactosidase_Abundance.pdf" and Oligosaccharide Abundance_by_Sialidase_Abundance.pdf"

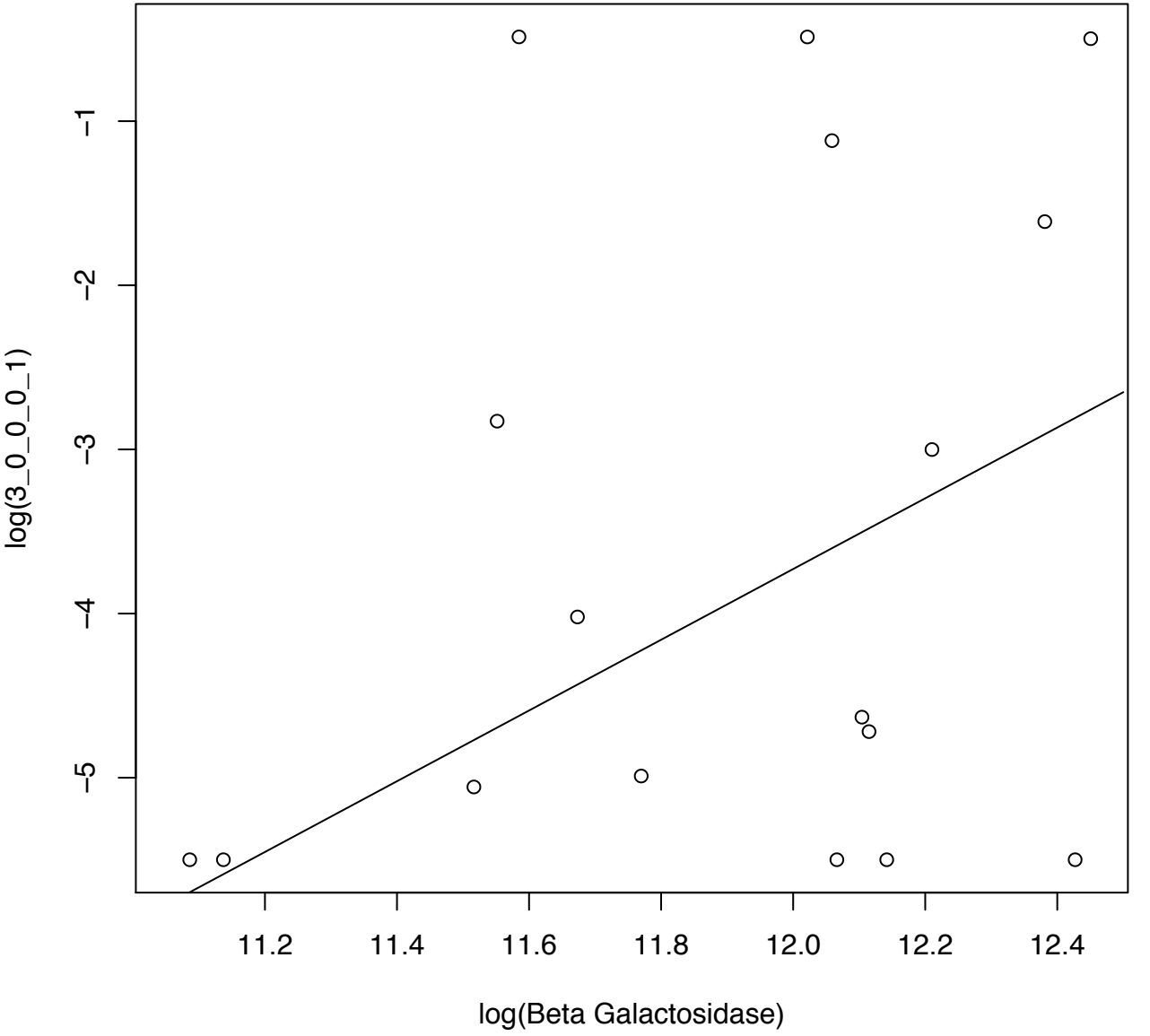
The x-axis shows log(enzyme), and the y-axis shows log(oligosaccharide). Circles represent observed data, and the solid line shows the fit from the tobit model.

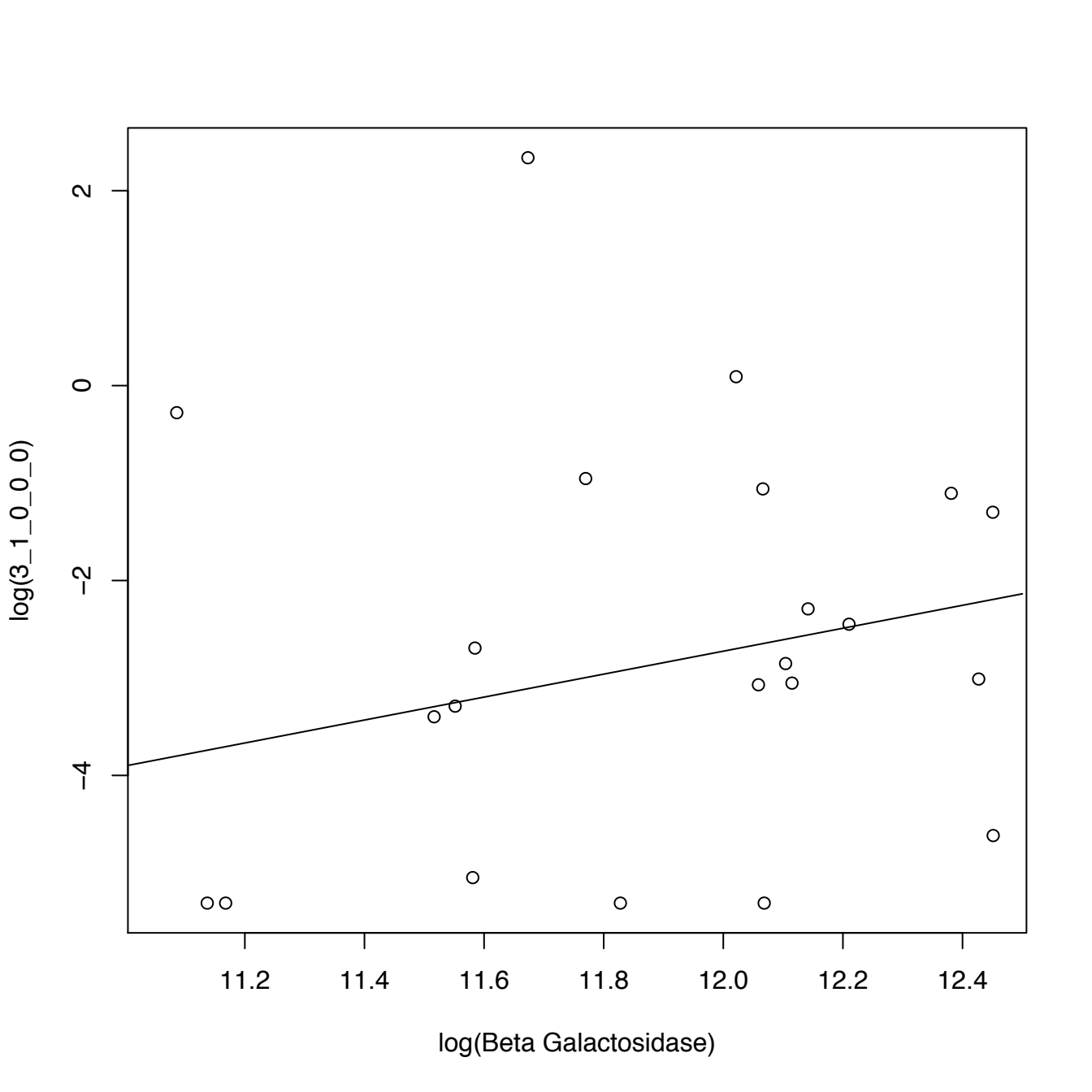

log(1_1_0_1_0)
−1
−2
−3
−4
−5
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

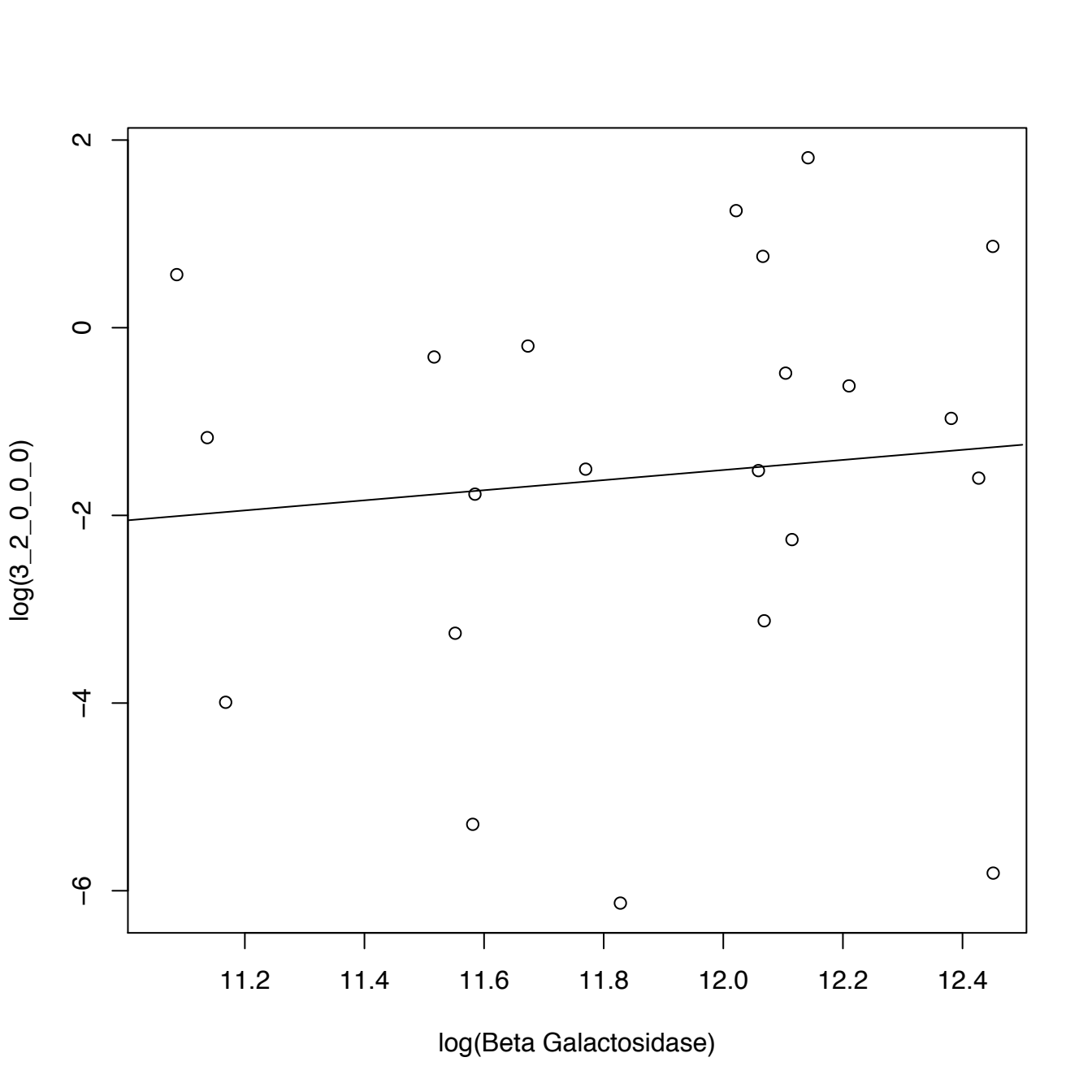

log(2_0_0_1_0)
0.5
0.0
−0.5
−1.0
−1.5
−2.0
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

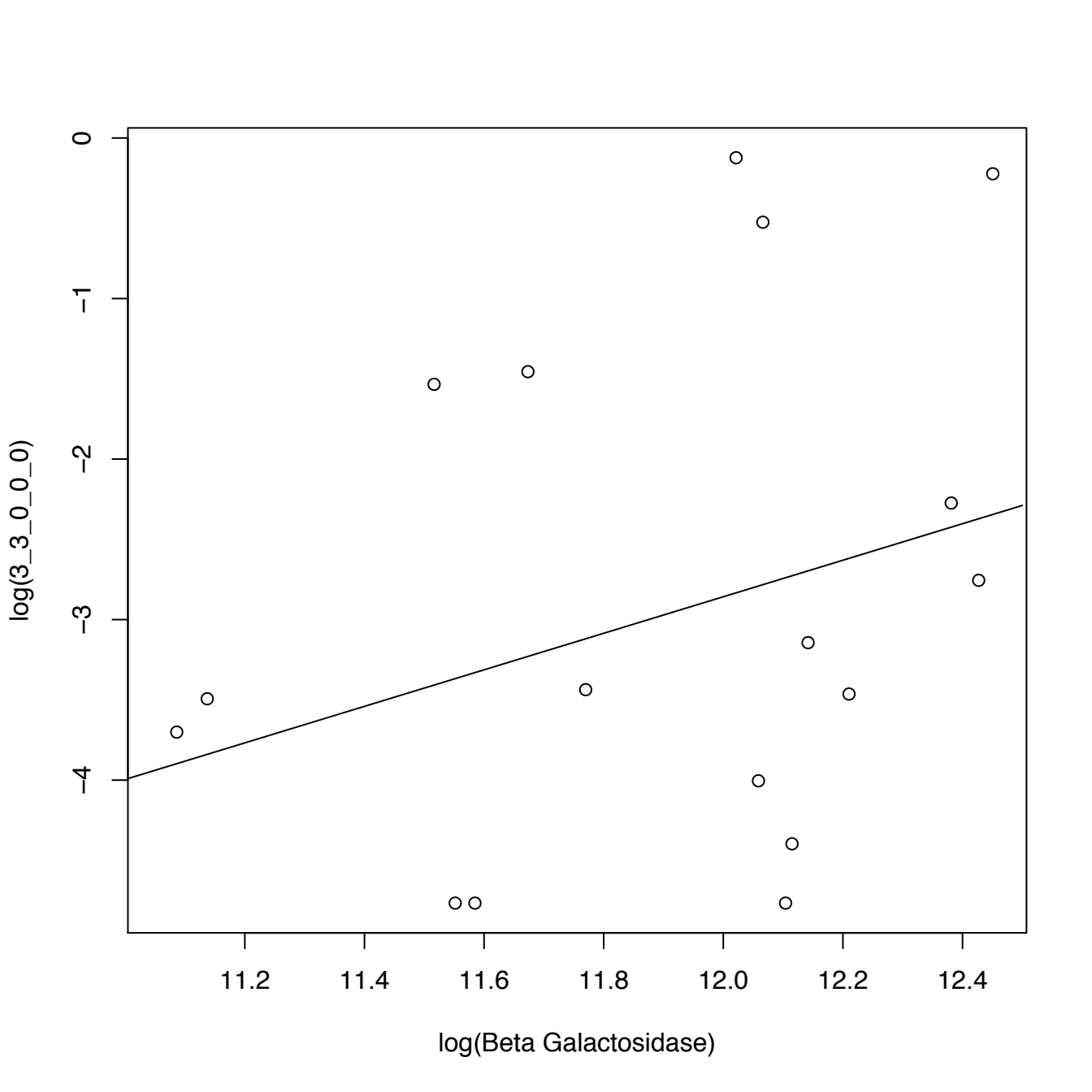

log(2_1_0_0_0)
0
−1
−2
−3
−4
−5
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

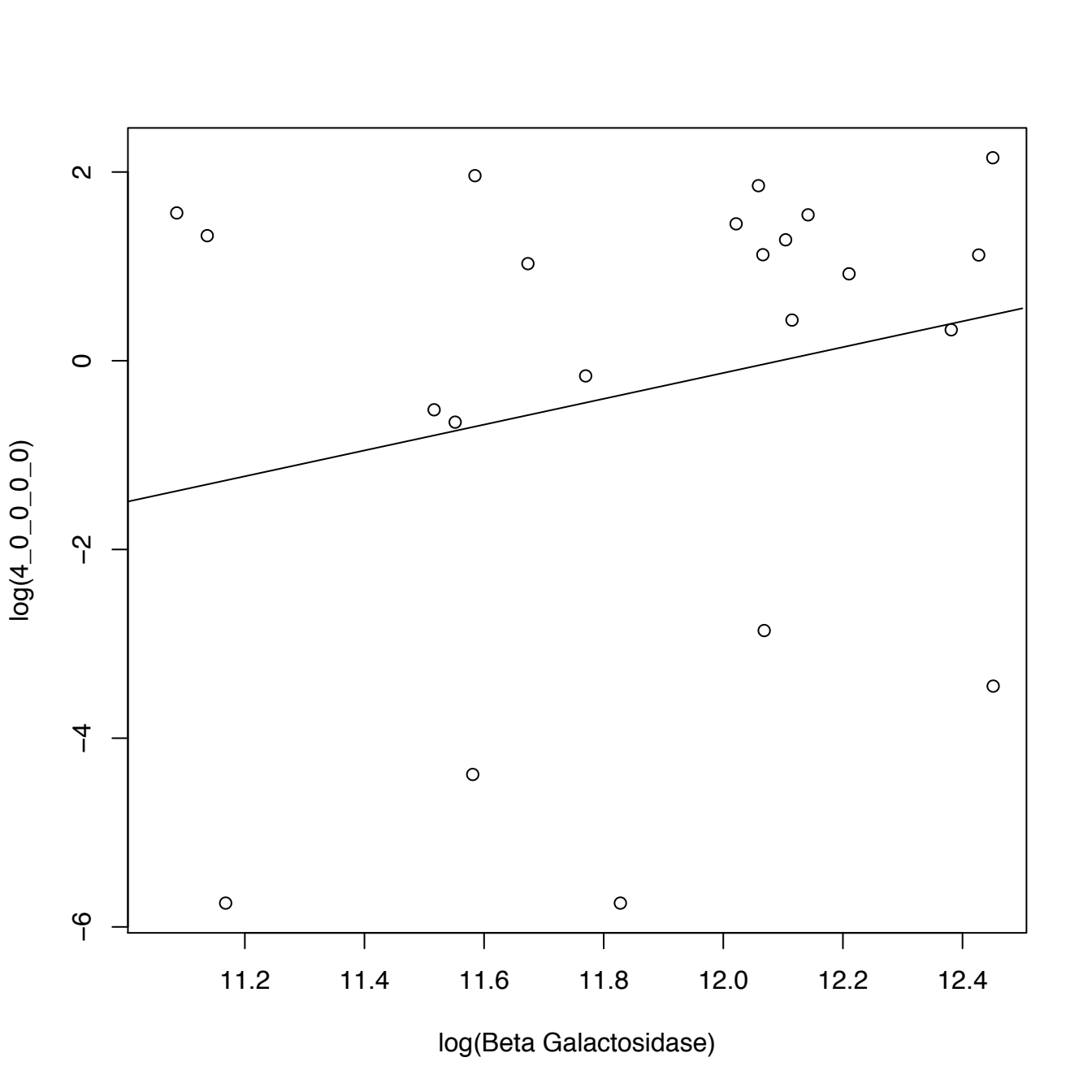

log(3_0_0_0_0)
2
0
−2
−4
−6
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

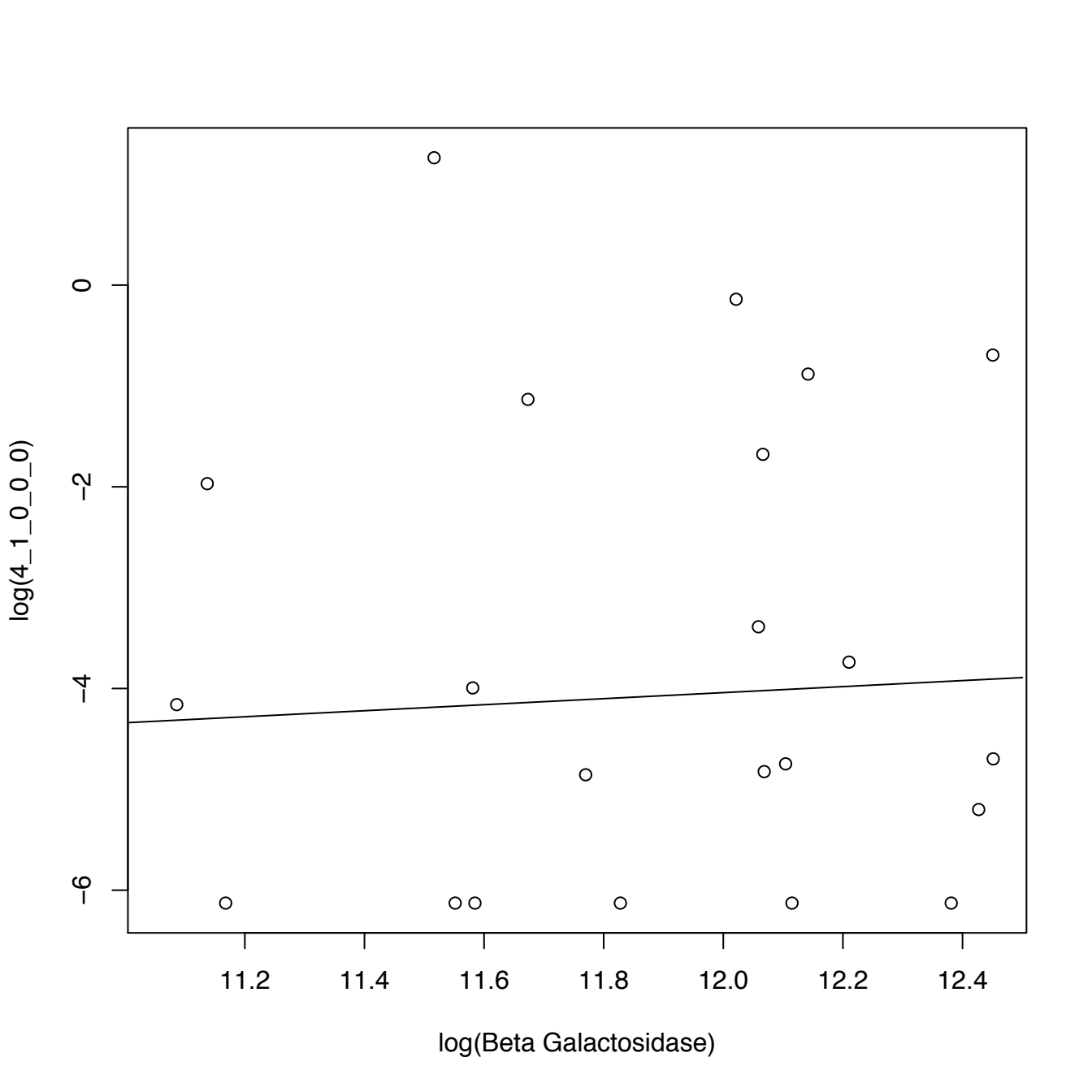

log(3_0_0_0_1)
−1
−2
−3
−4
−5
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

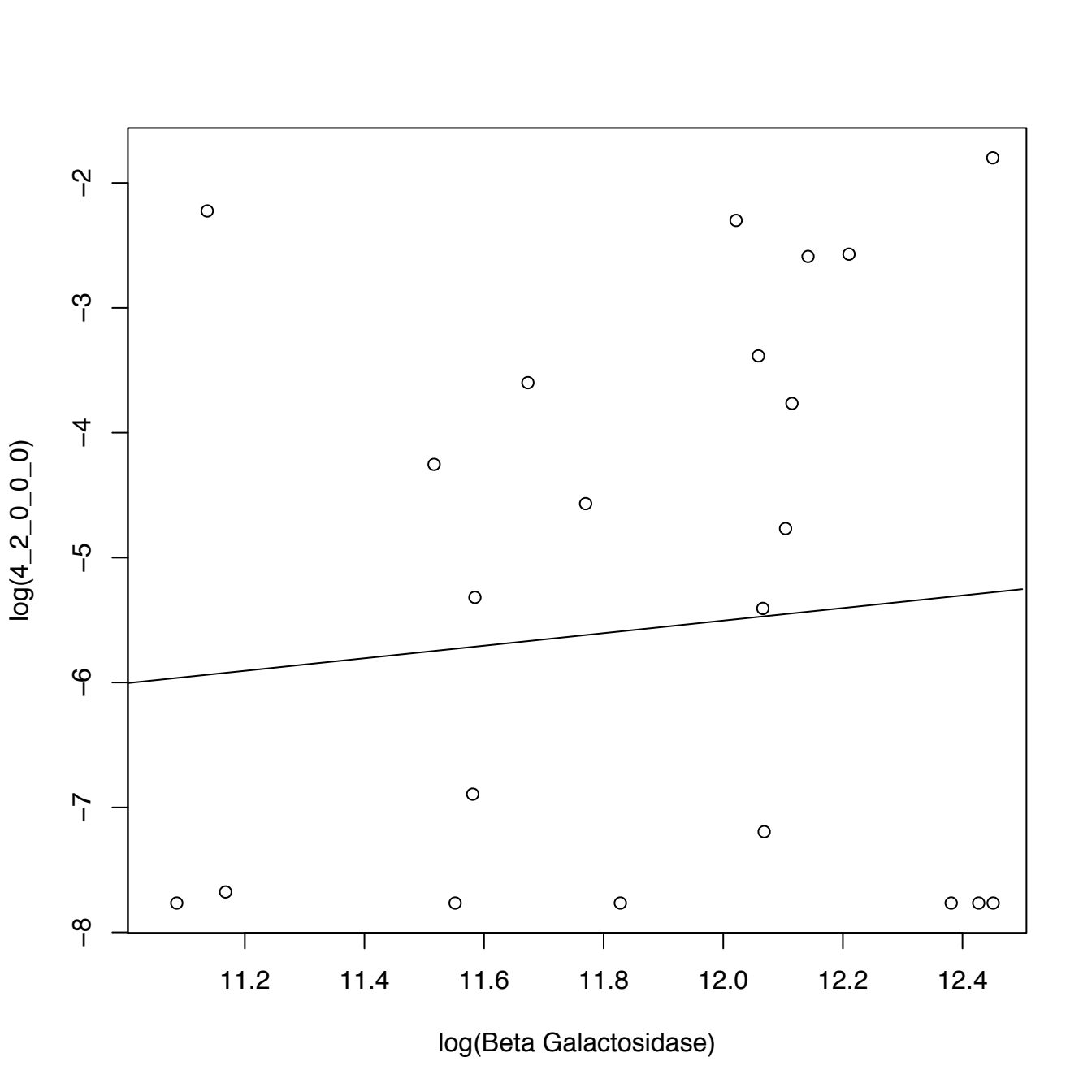

log(3_1_0_0_0)
2
0
−2
−4
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

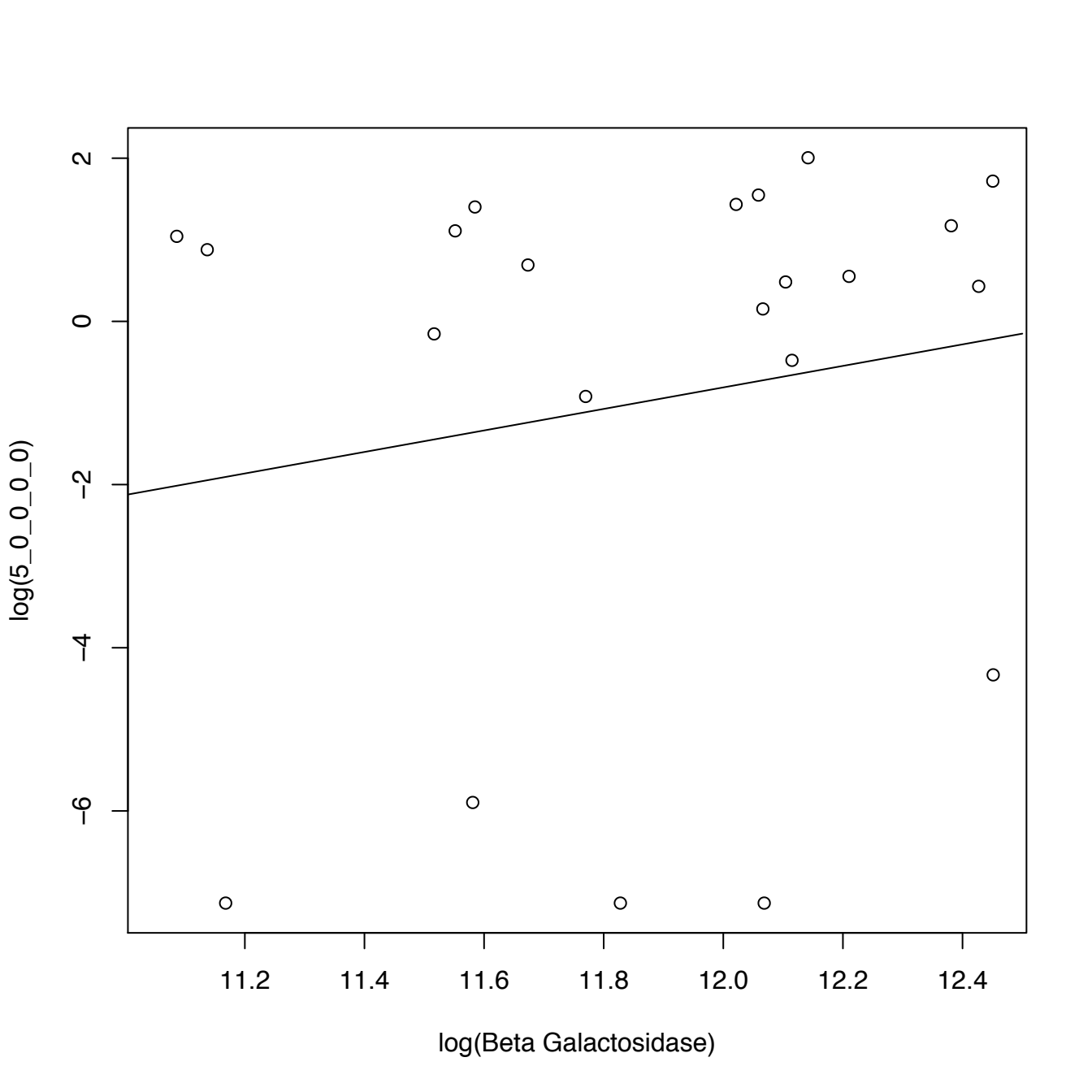

log(3_2_0_0_0)
2
0
−2
−4
−6
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

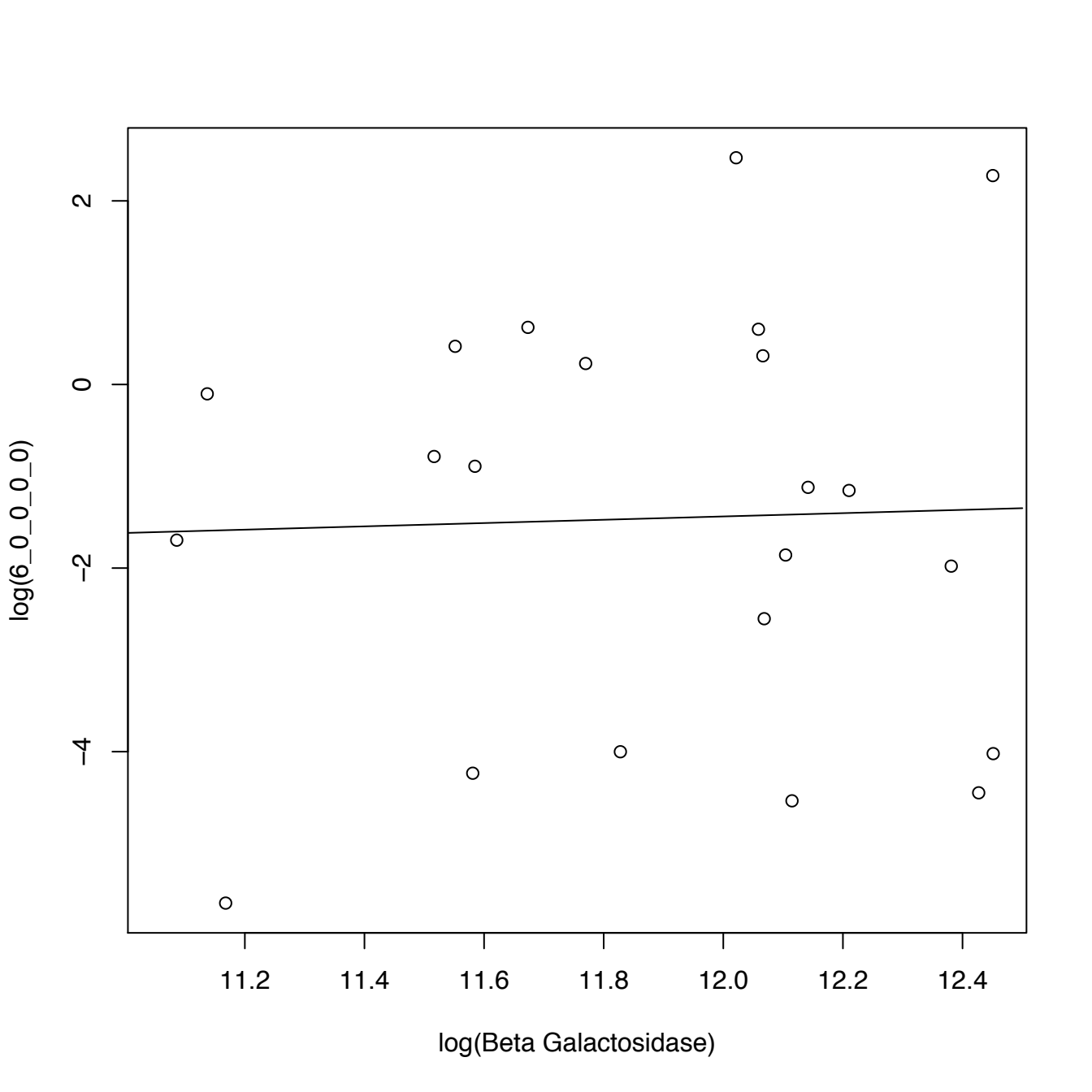

log(3_3_0_0_0)
0
−1
−2
−3
−4
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

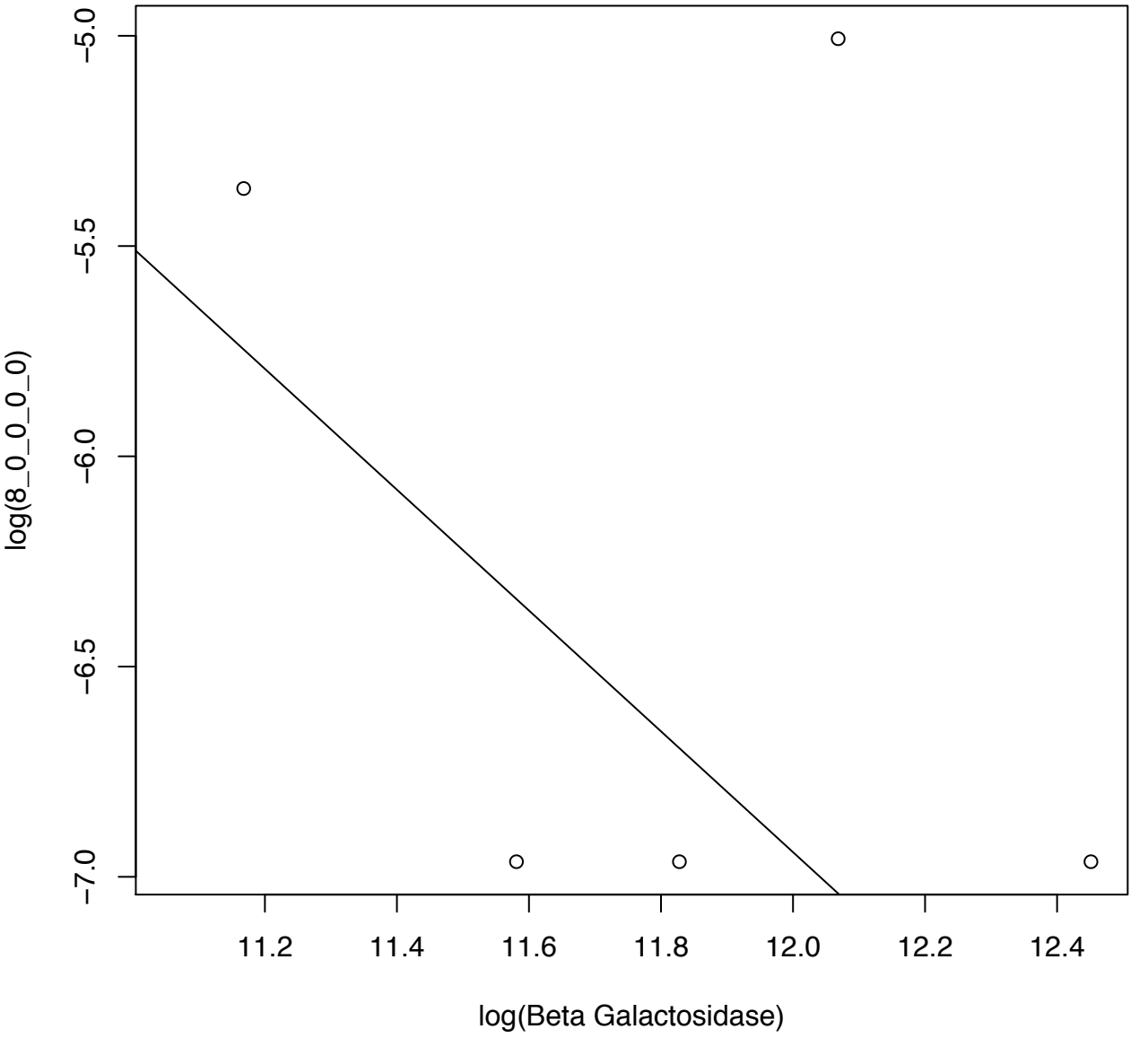

log(4_0_0_0_0)
2
0
−2
−4
−6
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

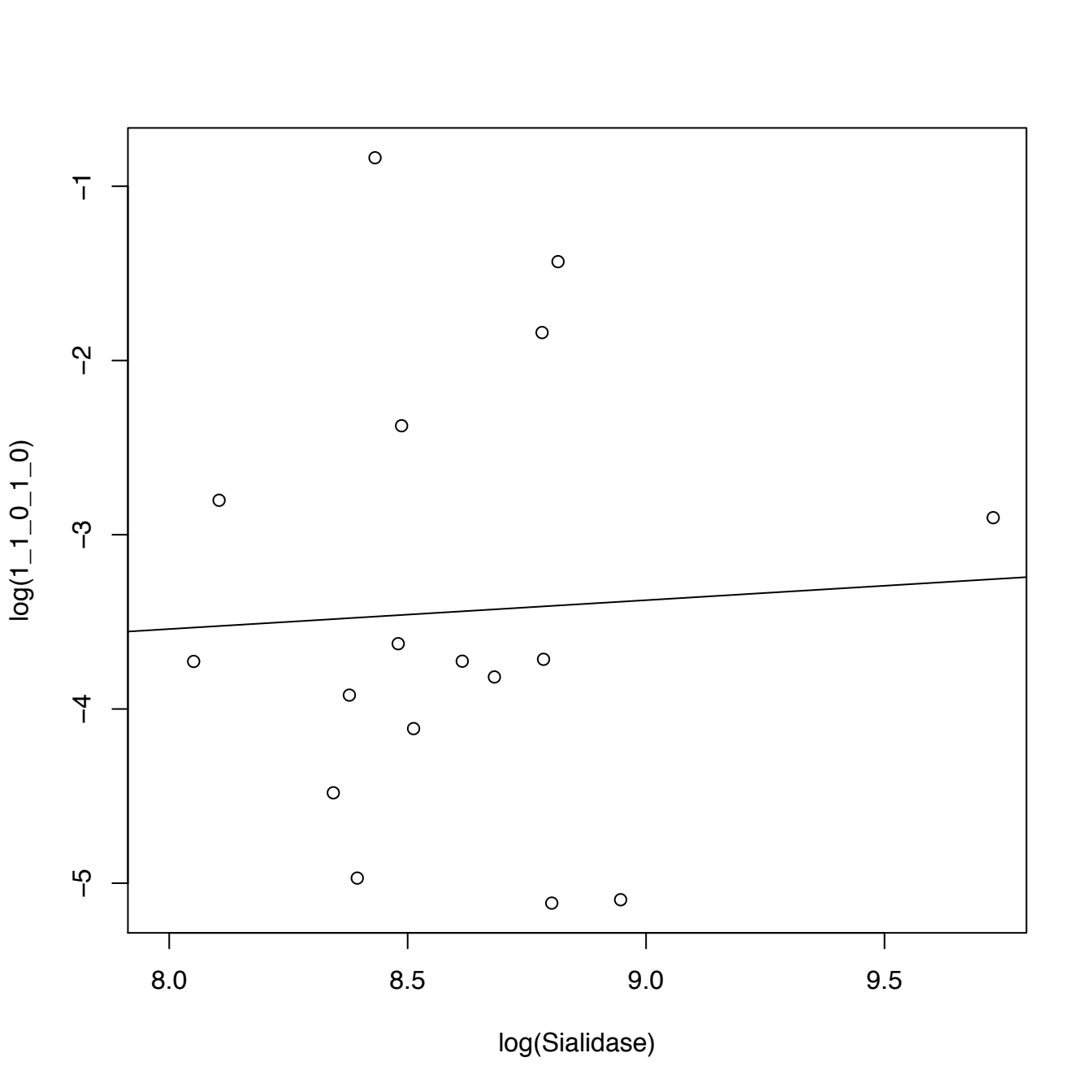

log(4_1_0_0_0)
0
−2
−4
−6
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

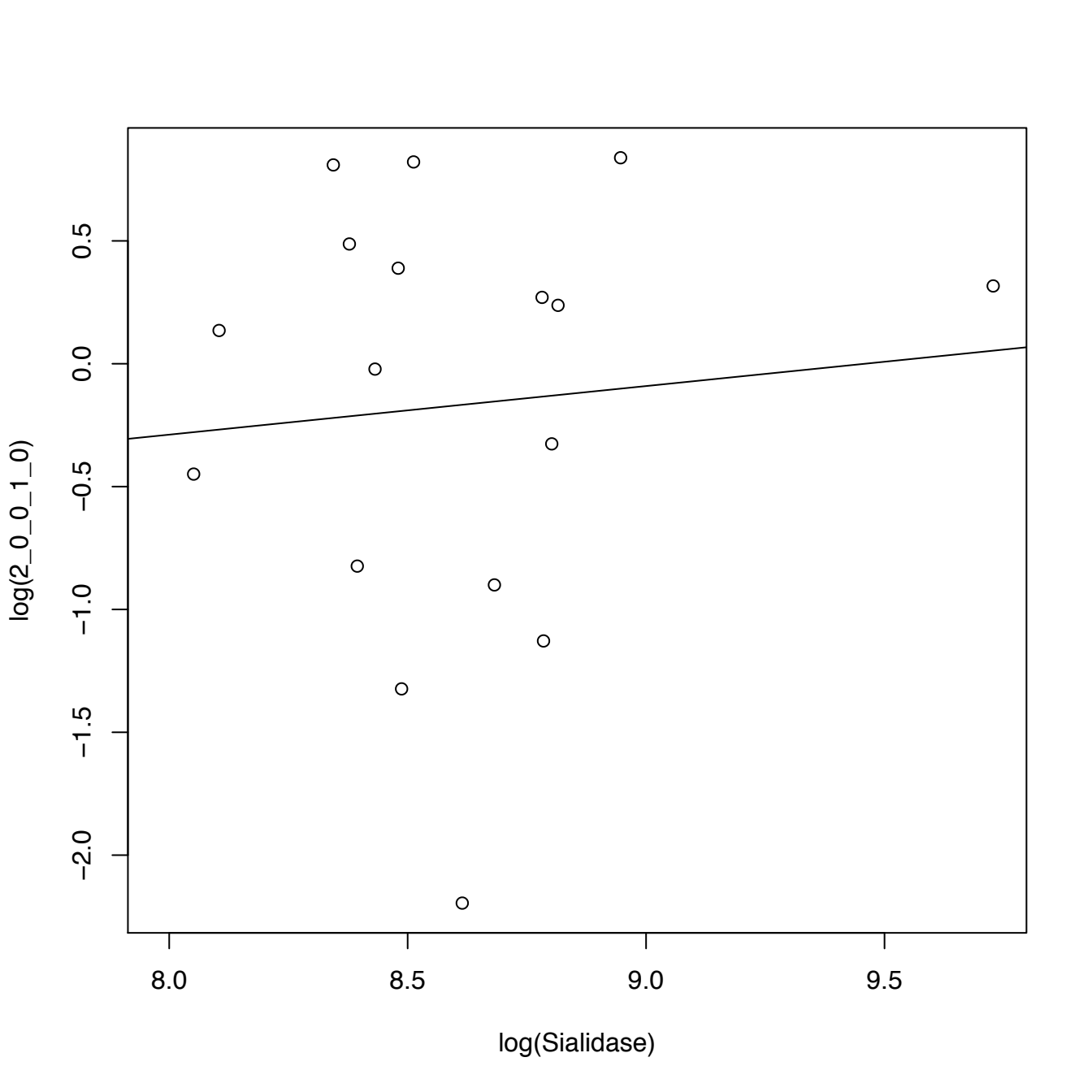

log(4_2_0_0_0)
−2
−3
−4
−5
−6
−7
−8
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

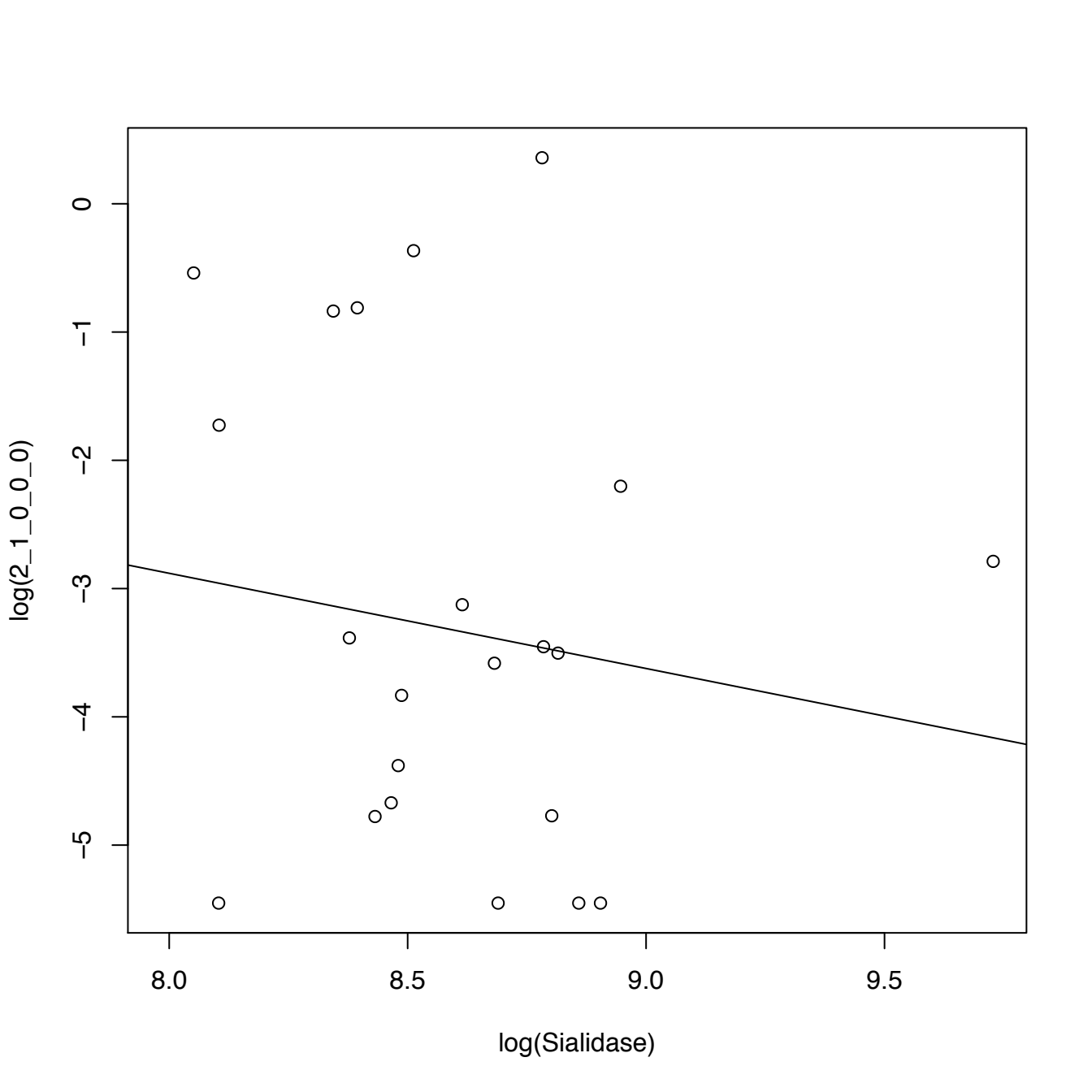

log(5_0_0_0_0)
2
0
−2
−4
−6
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

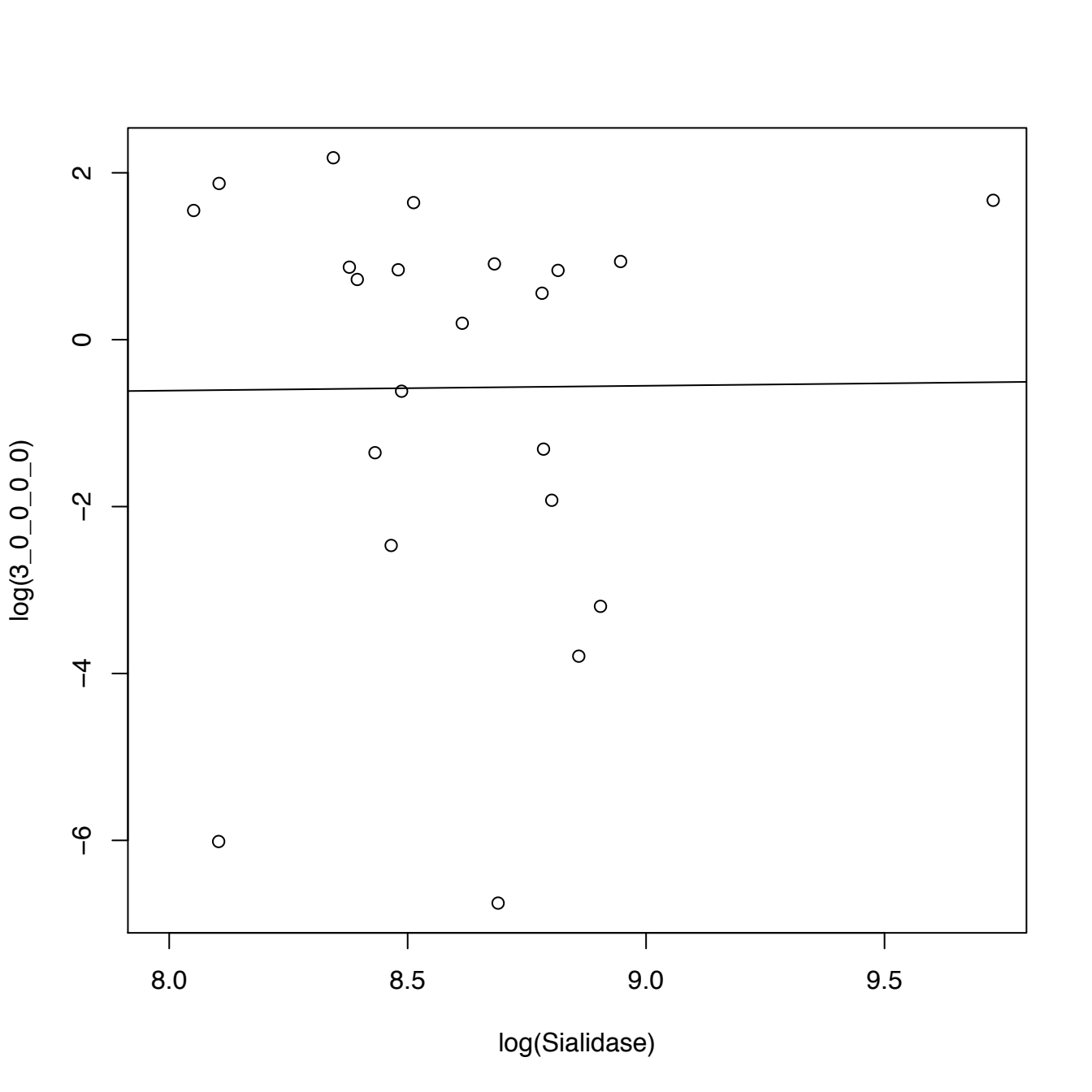

log(6_0_0_0_0)
2
0
−2
−4
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

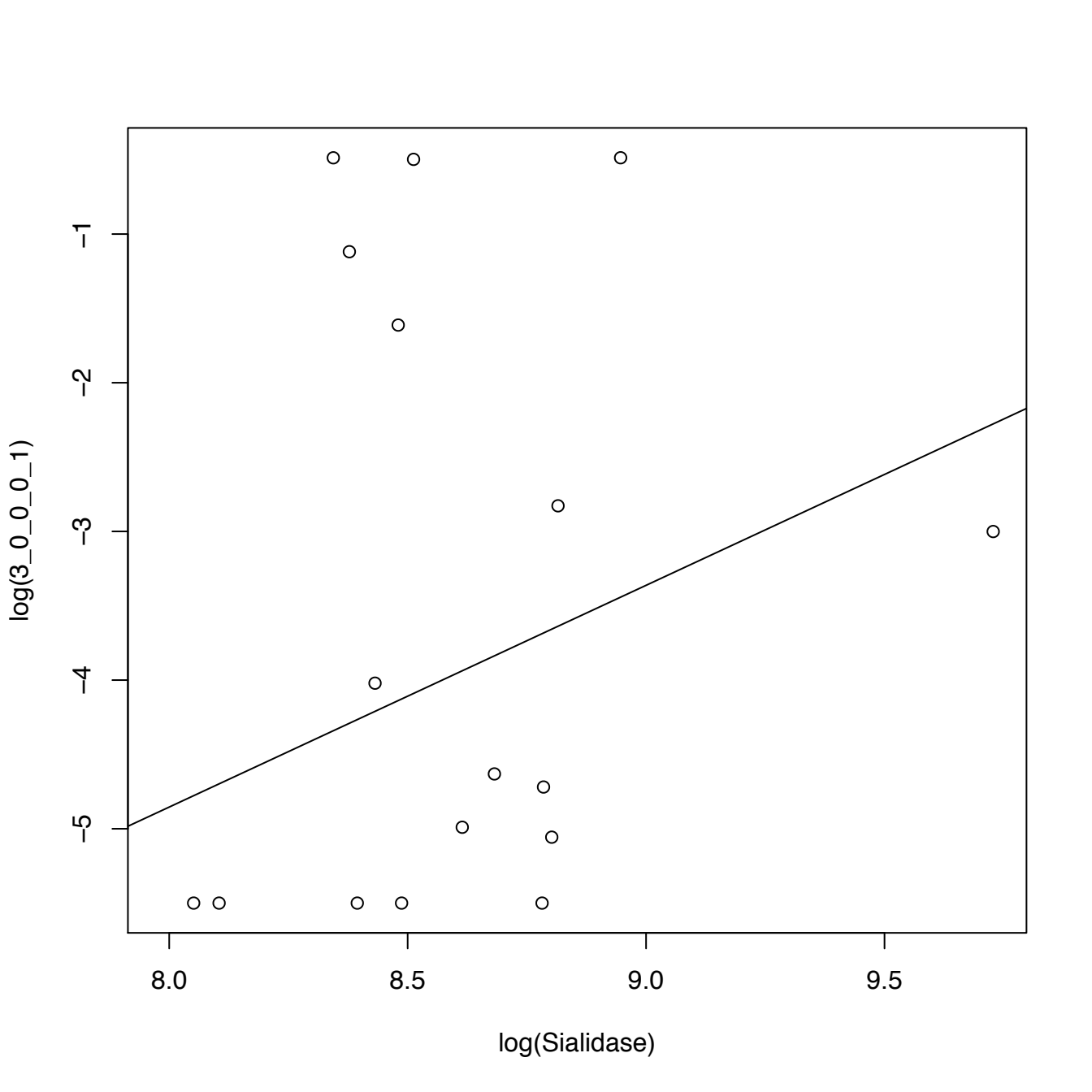

−5.0
−5.5
−6.0
−6.5
−7.0
log(8_0_0_0_0)
11.2
11.4
11.6
11.8
12.0
12.2
12.4
log(Beta Galactosidase)

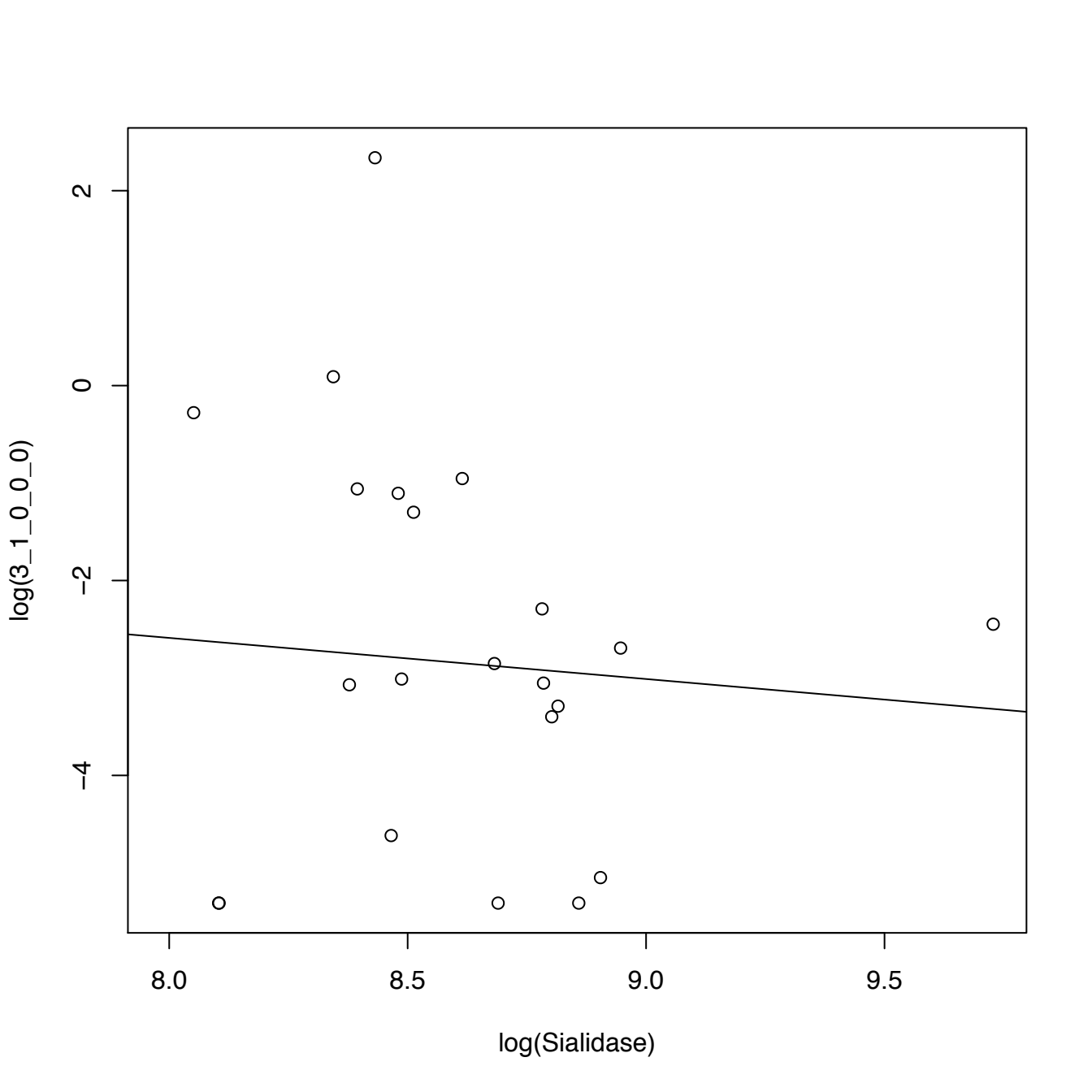

log(1_1_0_1_0)
−1
−2
−3
−4
−5
8.0
8.5
9.0
9.5
log(Sialidase)

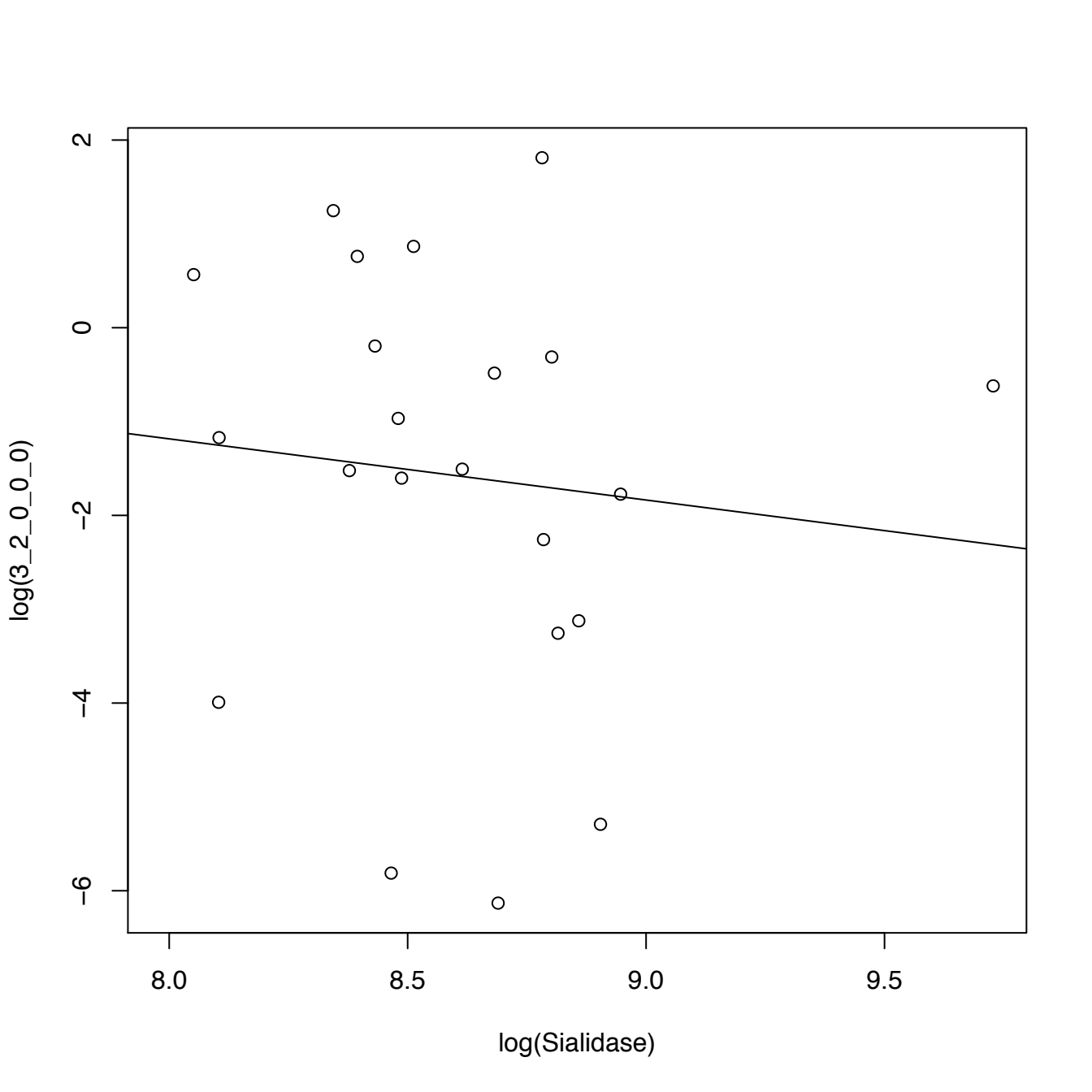

log(2_0_0_1_0)
0.5
0.0
−0.5
−1.0
−1.5
−2.0
8.0
8.5
9.0
9.5
log(Sialidase)

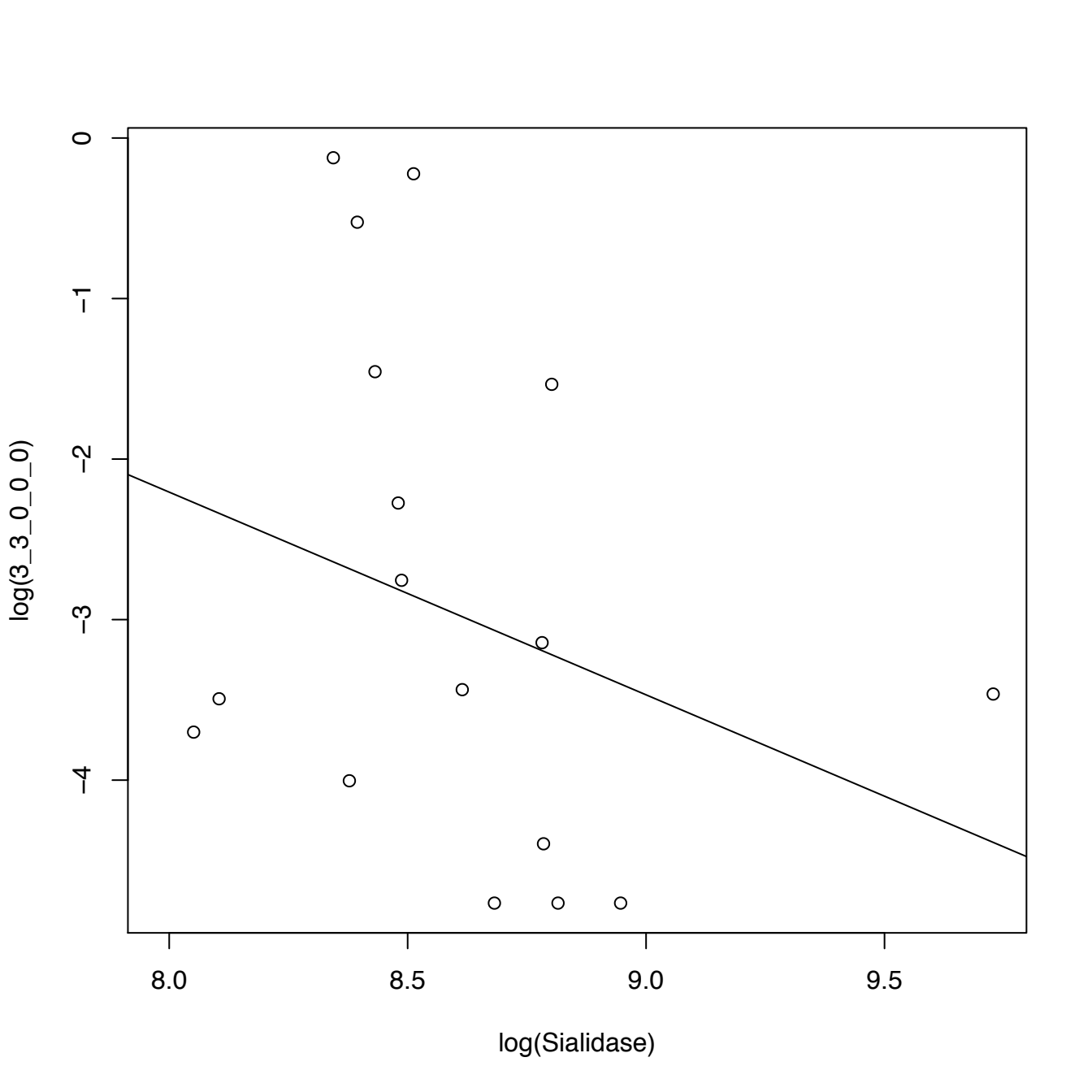

log(2_1_0_0_0)
0
−1
−2
−3
−4
−5
8.0
8.5
9.0
9.5
log(Sialidase)

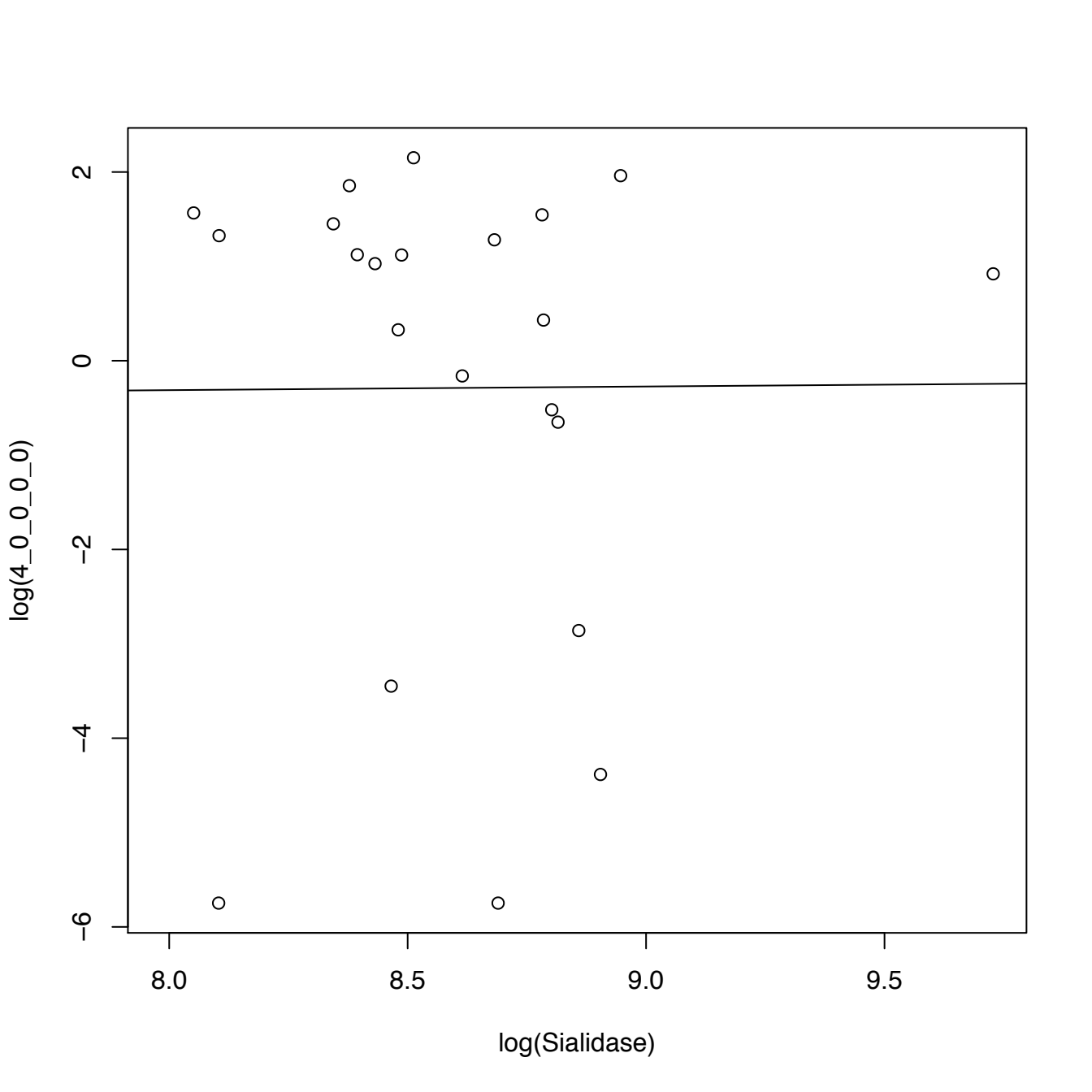

log(3_0_0_0_0)
2
0
−2
−4
−6
8.0
8.5
9.0
9.5
log(Sialidase)

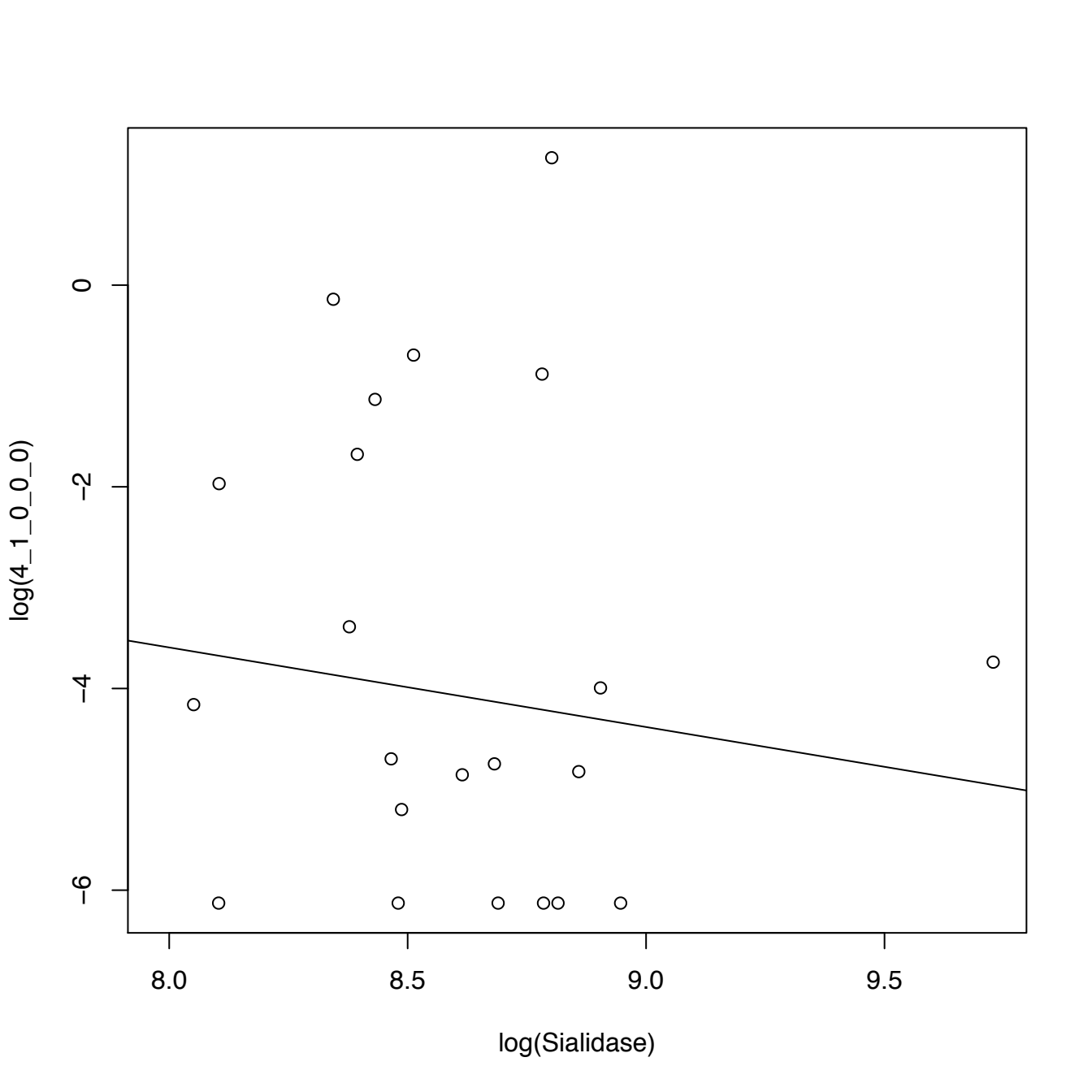

log(3_0_0_0_1)
−1
−2
−3
−4
−5
8.0
8.5
9.0
9.5
log(Sialidase)

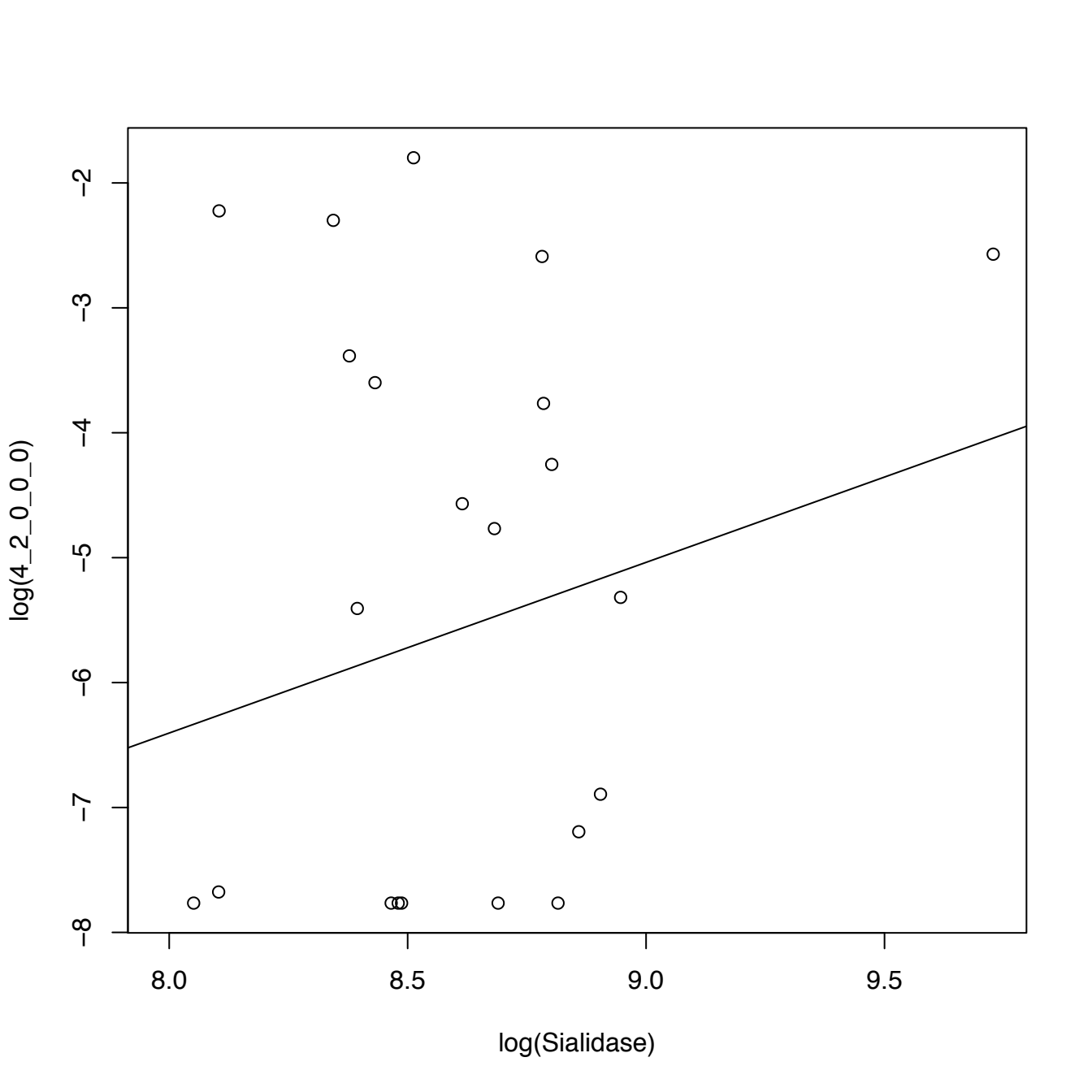

log(3_1_0_0_0)
2
0
−2
−4
8.0
8.5
9.0
9.5
log(Sialidase)

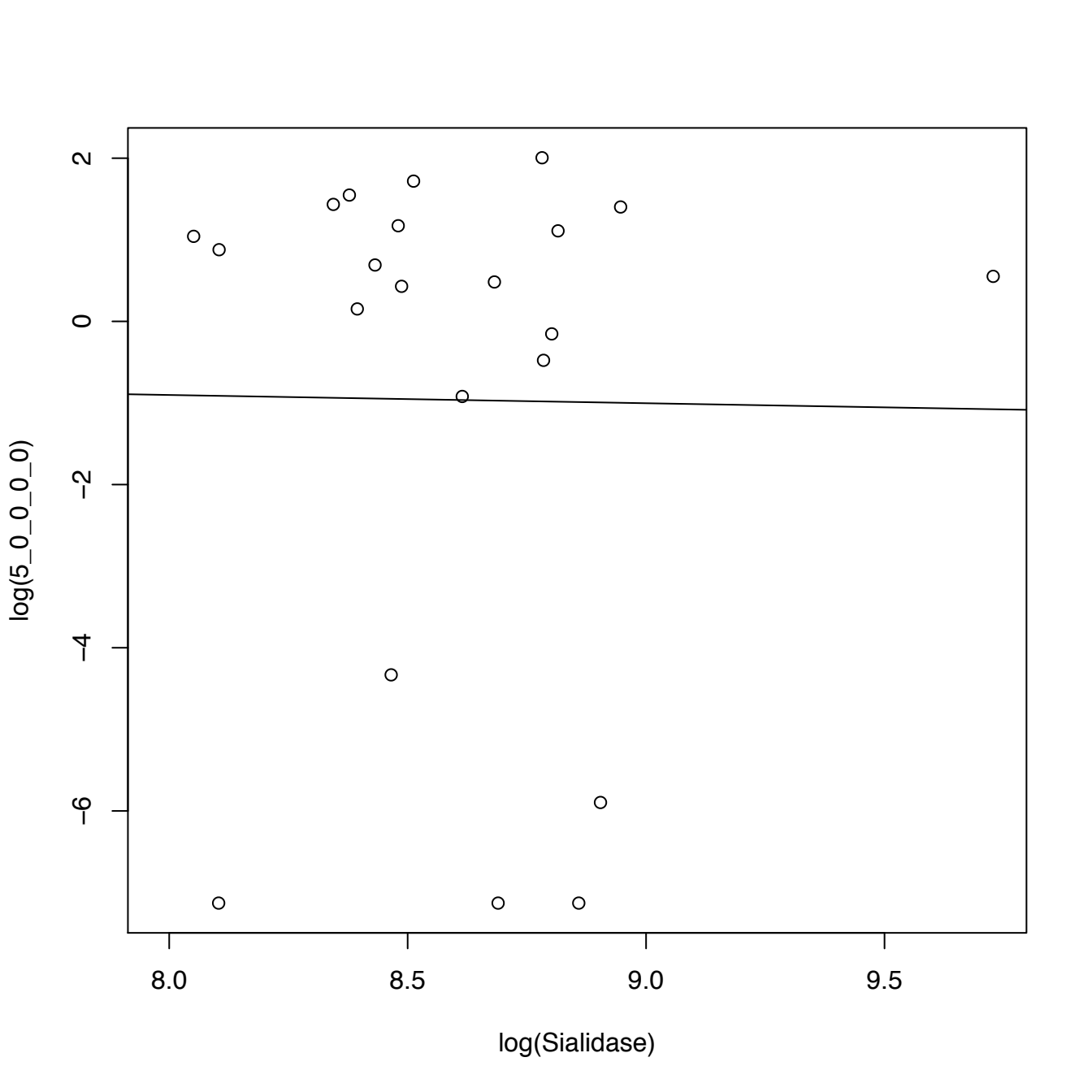

log(3_2_0_0_0)
2
0
−2
−4
−6
8.0
8.5
9.0
9.5
log(Sialidase)

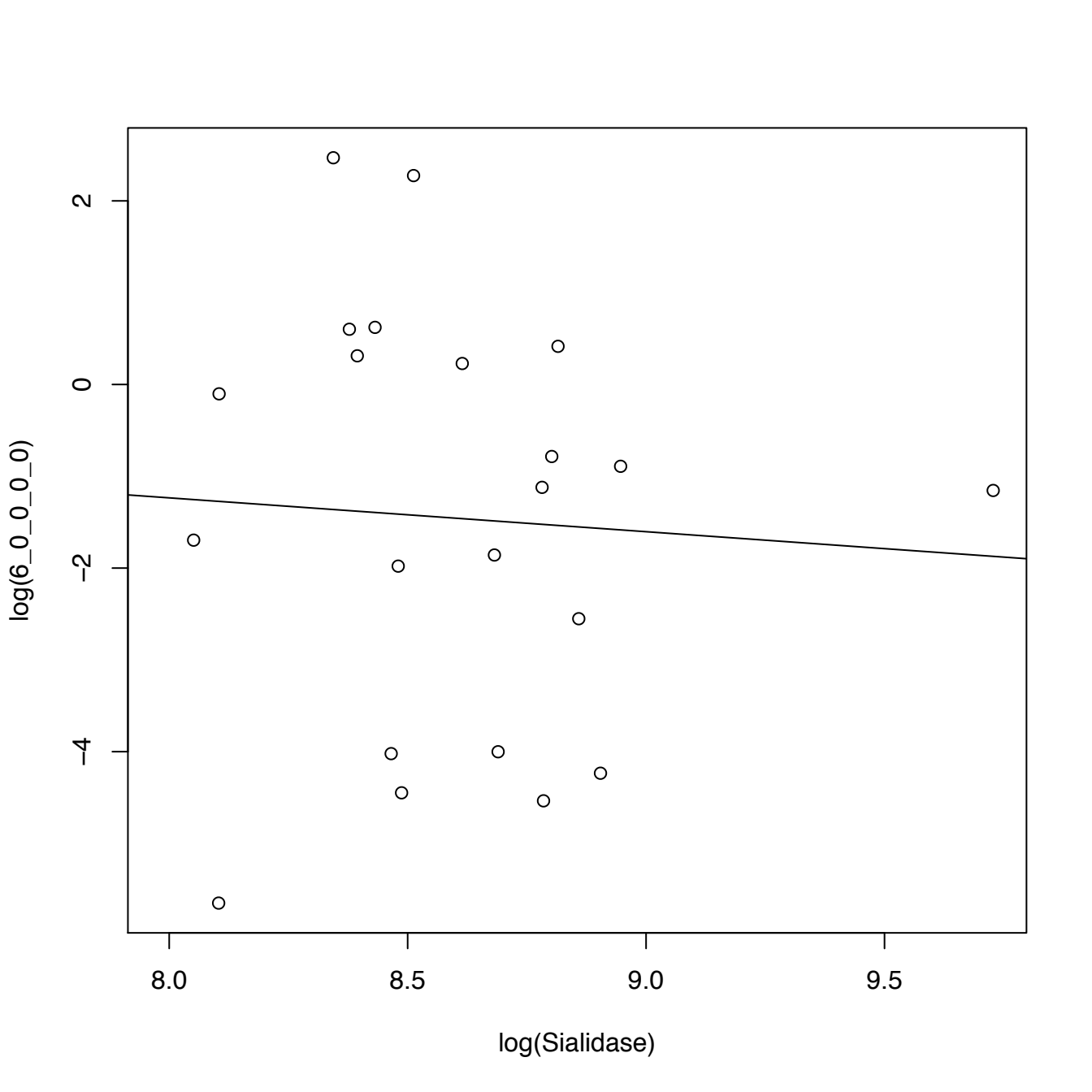

log(3_3_0_0_0)
0
−1
−2
−3
−4
8.0
8.5
9.0
9.5
log(Sialidase)

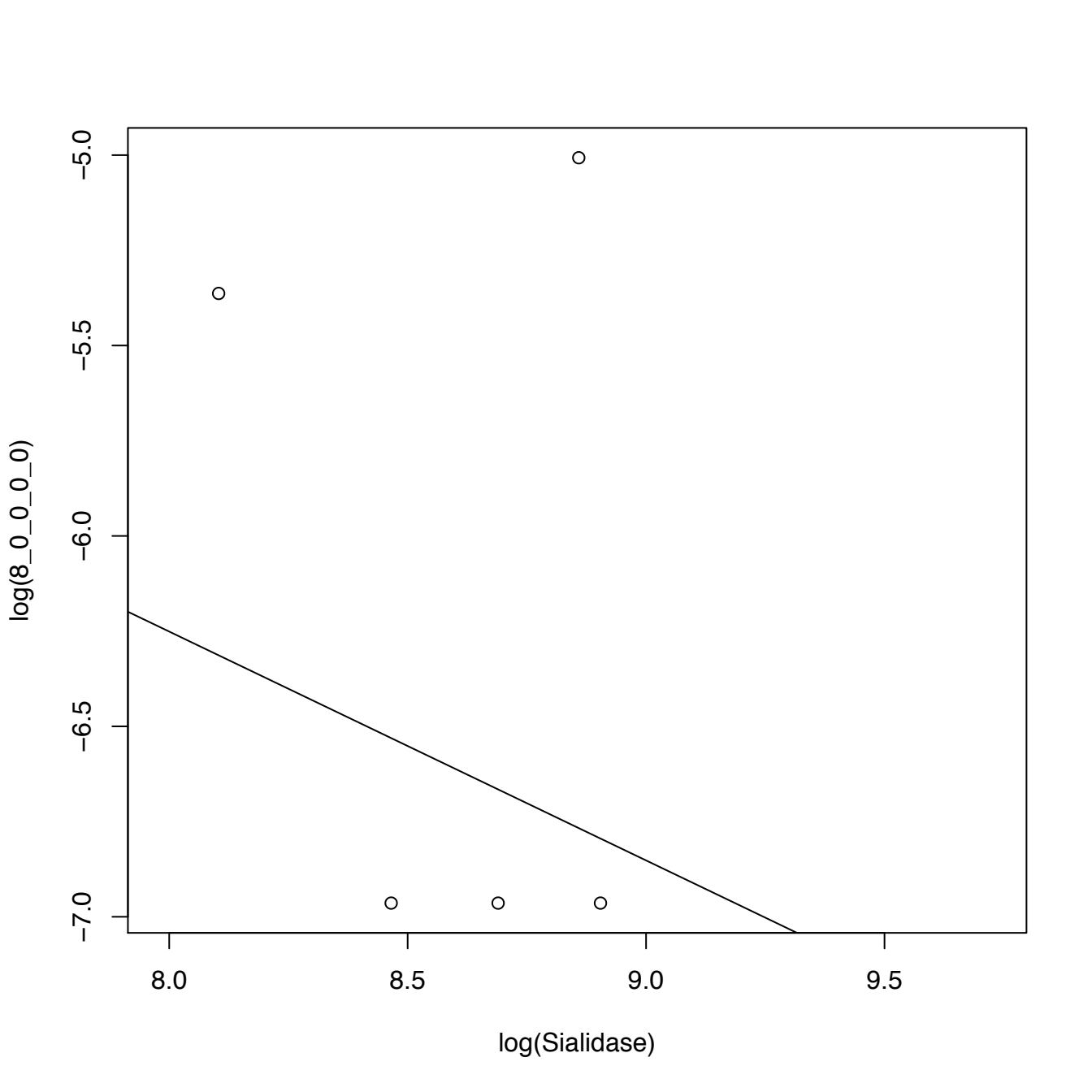

log(4_0_0_0_0)
2
0
−2
−4
−6
8.0
8.5
9.0
9.5
log(Sialidase)


log(4_1_0_0_0)
0
−2
−4
−6
8.0
8.5
9.0
9.5
log(Sialidase)


log(4_2_0_0_0)
−2
−3
−4
−5
−6
−7
−8
8.0
8.5
9.0
9.5
log(Sialidase)


log(5_0_0_0_0)
2
0
−2
−4
−6
8.0
8.5
9.0
9.5
log(Sialidase)


log(6_0_0_0_0)
2
0
−2
−4
8.0
8.5
9.0
9.5
log(Sialidase)


log(8_0_0_0_0)
−5.0
−5.5
−6.0
−6.5
−7.0
8.0
8.5
9.0
9.5
log(Sialidase)