Report

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1.Summary

2.Introduction

Uterotropic bioassay is one approach for screening chemicals for endocrine disrupting effect. An international mutilaboratory study was conducted with this method using known estrogen agonist(`EE`) and antagonist(`ZM`). The main goal is to assess whether the results were consistent across the laboratories. For one aspect, we care about:1)whether the method is successful for identifying endocrine disrupting effect of `EE` and `ZM`; 2)how dose response? is there a "change" dose level of `EE` for endocrine disrupting effect; 3)do the protocols differ in their sensitivity to detect estrogenic and anti-estrogenic effects; which one is recommended? For the other aspect, we care about:1) whether the results for abovementioned problems vary across labs;2)which labs stand out for being different?

The dataset including: response variable `uterus`(Uterus weight (mg)); key explanatory variables: `EE`(Dose of estrogen agonist,mg/kg/day),`ZM`(Dose of estrogen antagonist, mg/kg/day);other explanatory variables: `Lab`(19 Laboratory at which assay was conducted), `Protocol`(4 levels factor, representing immature female rats dosed by oral gavage (3 days), immature female rats dosed by injection (3 days), adult ovariectomized female rats dosed by injection (3 days), adult ovariectomized female rats dosed by injection (7 days) seperately.); `Weight`(Body weight of rat (g)),`Group` (Lab replicate group (6 rats were used per group)).

The characteristics of the dataset.

```
##1)We can consider `weight` and `uterus` as continuous variable. All other variables only have seperate
summary(bioassay)
```

```
##      uterus           weight            EE               ZM
##  Min.   : 10.4   Min.   : 27.7   Min.   : 0.000   Min.   :0.0000
##  1st Qu.: 32.8   1st Qu.: 48.4   1st Qu.: 0.010   1st Qu.:0.0000
##  Median : 80.0   Median : 56.1   Median : 0.300   Median :0.0000
##  Mean   :100.8   Mean   :106.7   Mean   : 1.875   Mean   :0.1009
##  3rd Qu.:124.4   3rd Qu.:200.2   3rd Qu.: 3.000   3rd Qu.:0.0000
##  Max.   :468.3   Max.   :341.0   Max.   :10.000   Max.   :1.0000
##  NA's   :4       NA's   :2
##  protocol       lab           group
##  A:1032   Hatano  : 264   2      : 246
##  B: 792   InEnvTox: 264   5      : 246
##  C: 594   Nihon   : 264   6      : 246
##  D: 263   Mitsubis: 263   7      : 246
##           Citijapa: 198   8      : 246
##           Sumitomo: 198   9      : 246
##           (Other) :1230   (Other):1205
```

```
bioassay %>% select(EE) %>% table()
```

```
## .
##    0 0.01 0.03  0.1  0.3    1    3   10
##  486  234  239  246  246  246  738  246
```

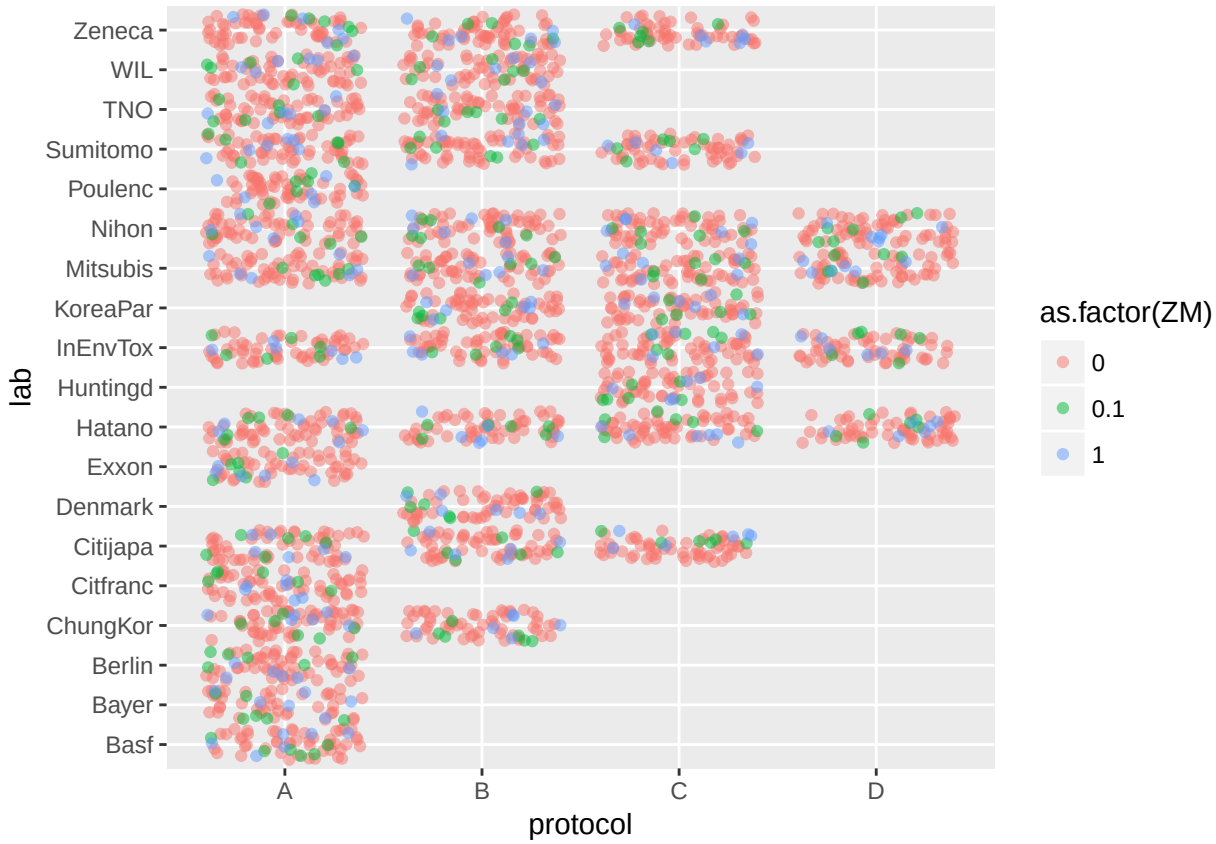
```
bioassay %>% select(ZM) %>% table()
```

```
## .
##    0  0.1    1
## 2189  246  246
```

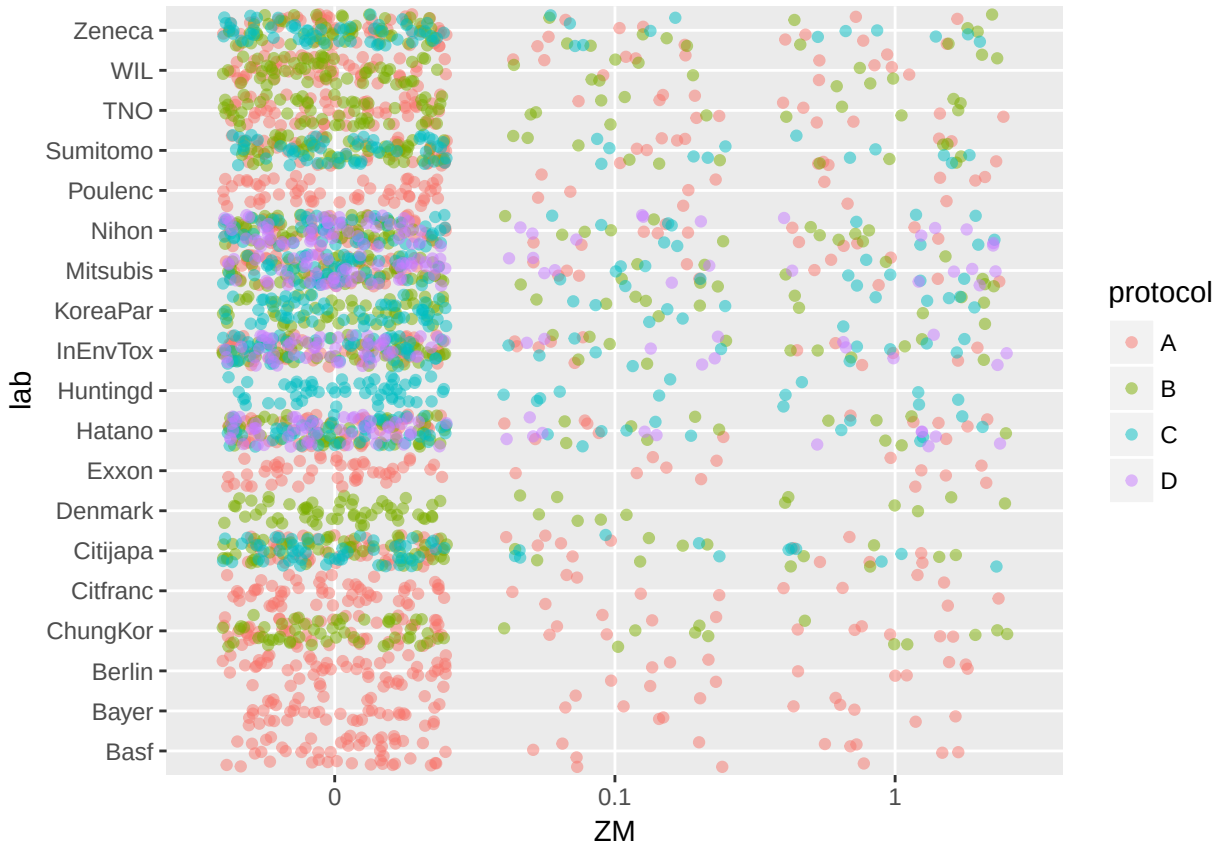
```
##2)There are significant interaction among variables. Because each labs adopted differet treatment.
table(bioassay$EE,bioassay.fac$lab)
```

```
##
##        Basf Bayer Berlin ChungKor Citfranc Citijapa Denmark Exxon Hatano
##   0      12    12     12       24       12       36      12     6     48
##   0.01    0     0      6       12        6       18       6     6     24
##   0.03    6     0      6       12        6       18       6     6     24
##   0.1     6     6      6       12        6       18       6     6     24
##   0.3     6     6      6       12        6       18       6     6     24
##   1       6     6      6       12        6       18       6     6     24
##   3      18    18     18       36       18       54      18    18     72
##   10      6     6      6       12        6       18       6     6     24
##
##        Huntingd InEnvTox KoreaPar Mitsubis Nihon Poulenc Sumitomo TNO WIL
##   0          12       48       24       48    48      12       36  24  24
##   0.01        6       24       12       24    24       6       18  12  12
##   0.03        6       24       12       23    24       6       18  12  12
##   0.1         6       24       12       24    24       6       18  12  12
##   0.3         6       24       12       24    24       6       18  12  12
##   1           6       24       12       24    24       6       18  12  12
##   3          18       72       36       72    72      18       54  36  36
##   10          6       24       12       24    24       6       18  12  12
##
##        Zeneca
##   0        36
##   0.01     18
##   0.03     18
##   0.1      18
##   0.3      18
##   1        18
##   3        54
##   10       18
```

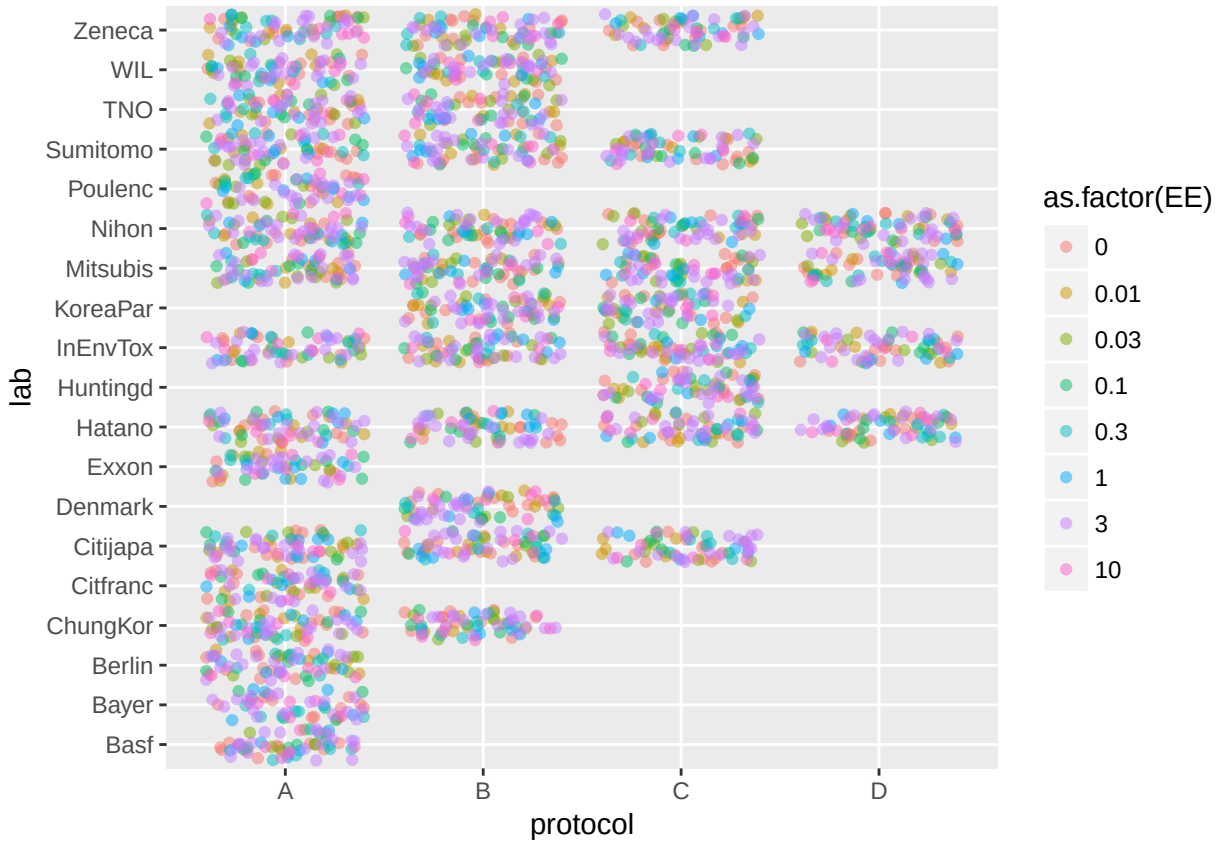
```
##visualization for points
ggplot(data=bioassay,mapping = aes(y = lab,x = protocol,color=as.factor(ZM)))+
  geom_jitter(alpha=0.5)
```



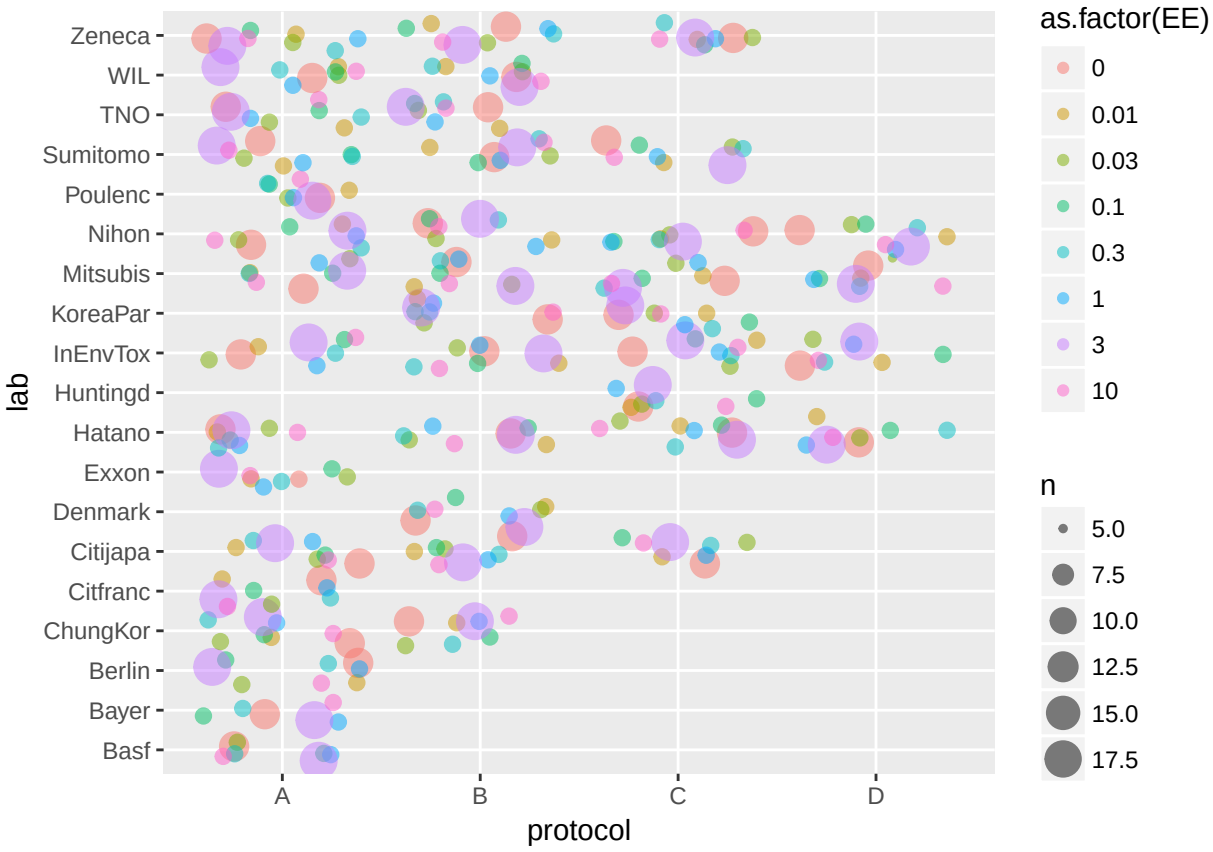

```
ggplot(data=bioassay.fac,mapping = aes(y = lab,x = ZM,color=protocol))+
  geom_jitter(alpha=0.5)
```




```
ggplot(data=bioassay,mapping = aes(y = lab,x = protocol,color=as.factor(EE)))+
  geom_jitter(alpha=0.5)
```




```
ggplot(data=bioassay,mapping = aes(y = lab,x = protocol,color=as.factor(EE)))+
     geom_count(alpha=0.5,position = "jitter")
```



3.Methods and Model

3.1 EDA

From the plot, we get three main information on the dataset.1)linear regession assumption.(quadratic form) 2)There are significant interaction among variables. Because each labs adopted differet treatment. So we have to include the iteraction into our regression model. 3)Many discrete variables. We need to keep the model simple.

I chose boxplot to illustrate the relationship between continuous variable (`uterus`) and discrete variable(`EE` and `ZM`).Different colors represent different `labs`, and I use facet to represent different `protocol`. From the plot1, we can see that: 1)The trend for expection looks like quadratic form; But overall, the uterus decrease as the zm increase; The variance under different level of `ZM` is different. The variance of `ZM`=0 is much larger than the other two groups; 2)Overall, the boxplots look similar. However, the different labs act different especially when `ZM`=0. Some labs, like TNO in `protocol="B",ZM=0.1` acts like an "outlier". There is interaction between `lab` and `ZM`. 3)Each labs adopted different treatment. So for some case, limited labs conducted the study. Similar for the case `EE`.

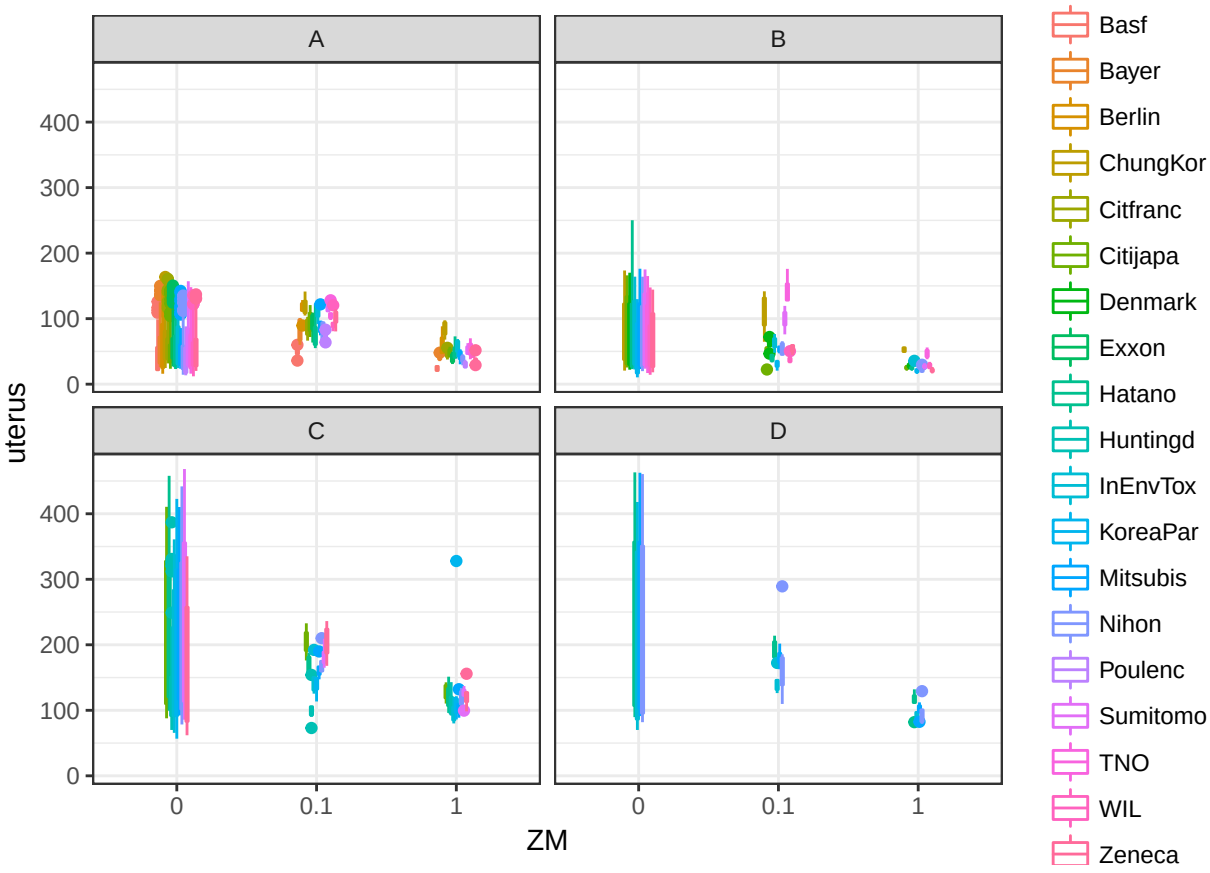
I chose scatterplots with regression line to illustrate the relationship between two continuous variable (`uterus` and `weight`). Because the significant differece on `weight` between immature rats and adult rats, I use facet to represent different `protocol`. Notice the range for y-axis is different. We can see that: 1)For immature rats, `weight` seems do not effect the `uterus`. While for adult rats, the `uterus` decrease as the `weight` increase. There is significant interaction.

```
##--------------------boxplot
##different labs have diff mean on ZM. interaction on labs and protocol.
##when consider ZM as numeric, the diff is not sig in pic. but in factor - diff.
```

```
###4var:ZM

ggplot(data=bioassay.fac,mapping = aes(y = uterus,x = ZM,color=lab))+
  geom_boxplot()+theme_bw()+facet_wrap(~ protocol)
```

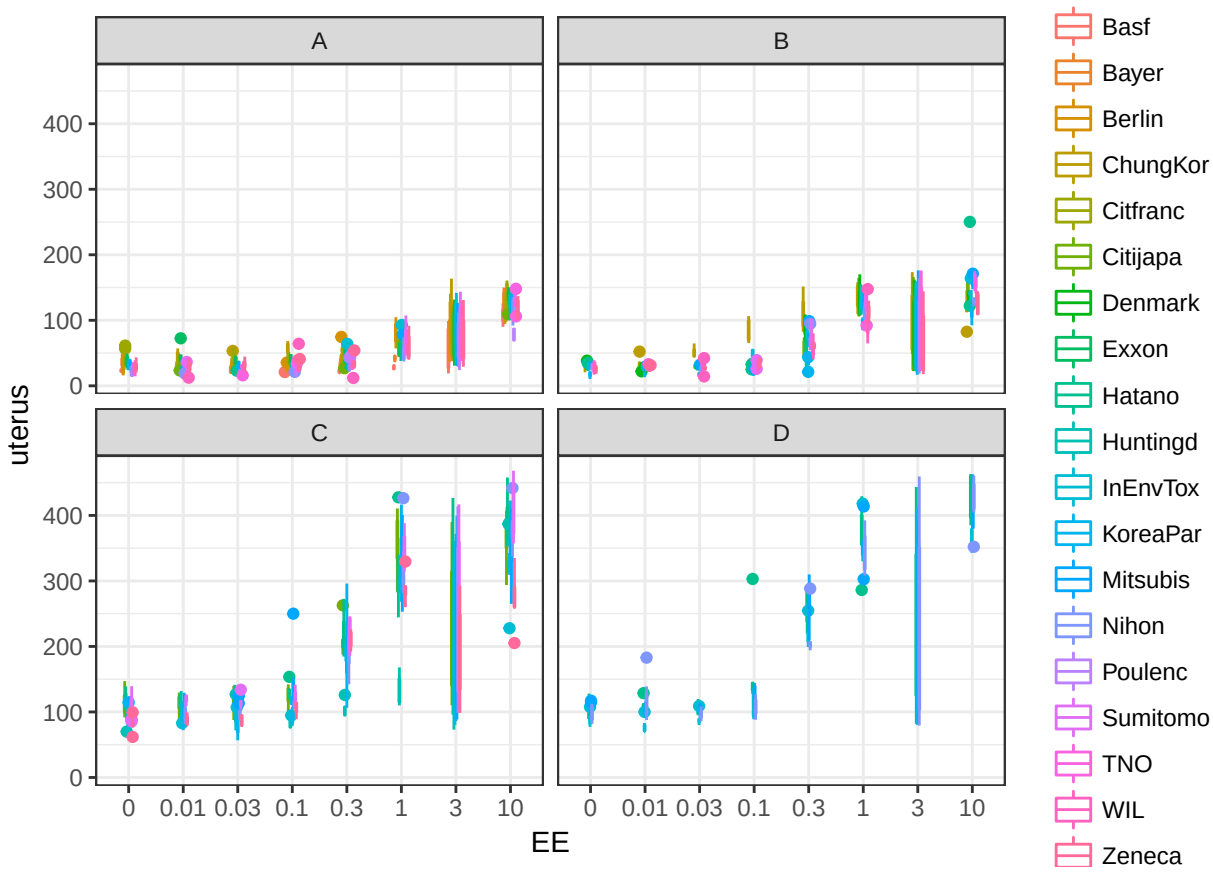
```
## Warning: Removed 4 rows containing non-finite values (stat_boxplot).
```




```
##caculate the mean for different group
#bioassay.fac %>% group_by(ZM) %>% summarize(mean=mean(uterus,na.rm=T))

##4var:EE
ggplot(data=bioassay.fac,mapping = aes(y = uterus,x = EE,color=lab))+
  geom_boxplot()+theme_bw()+facet_wrap(~ protocol)
```

```
## Warning: Removed 4 rows containing non-finite values (stat_boxplot).
```

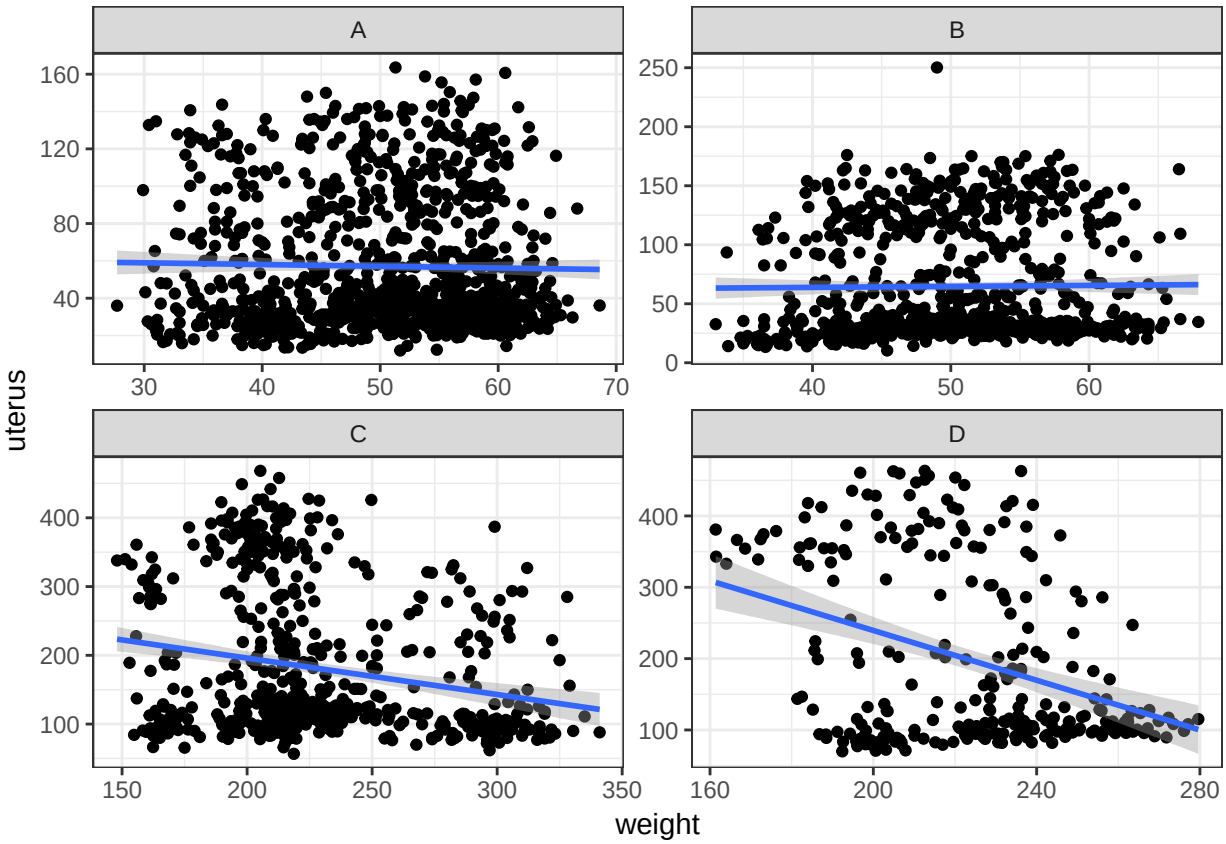



```
##caculate the mean for different group.
#bioassay.fac %>% group_by(EE) %>% summarize(mean=mean(uterus,na.rm=T))

##weights
ggplot(data=bioassay.fac,mapping = aes(y = uterus,x = weight))+
  geom_point()+geom_smooth(method = "lm")+theme_bw()+facet_wrap(~ protocol,scales = "free")
```

```
## Warning: Removed 4 rows containing non-finite values (stat_smooth).
```

```
## Warning: Removed 4 rows containing missing values (geom_point).
```



3.2 Model

I build a linear regression model with all variables except `group`. Apart from `weight` and `uterus`, I consider all other variables as factor. Also, I include the interaction between estrogen chemicals and different labs, protocols. Notice for the factors need to use a set of dummy variables to represent, so there are 218 predictors including the intercept.

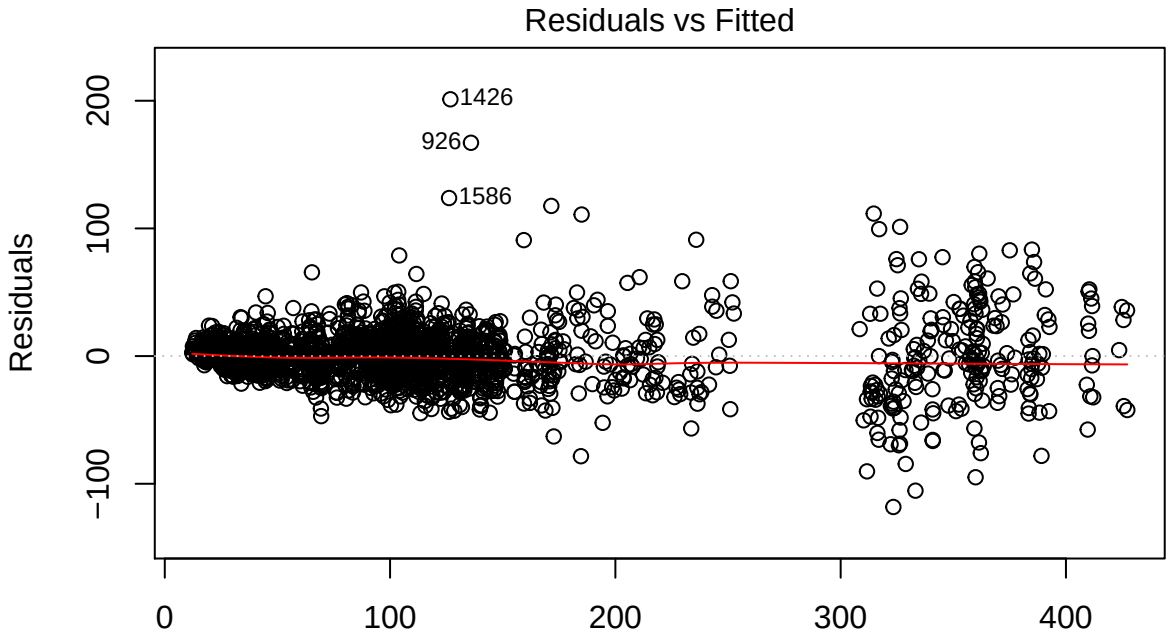
$Y = intercept + \beta_1 EE + \beta_2 ZM + \beta_3 lab + \beta_4 protocol + \beta_5 weight + \beta_6 EE : protocol + \beta_7 ZM : protocol + \beta_8 EE : lab + \beta_9 ZM : lab + \epsilon$

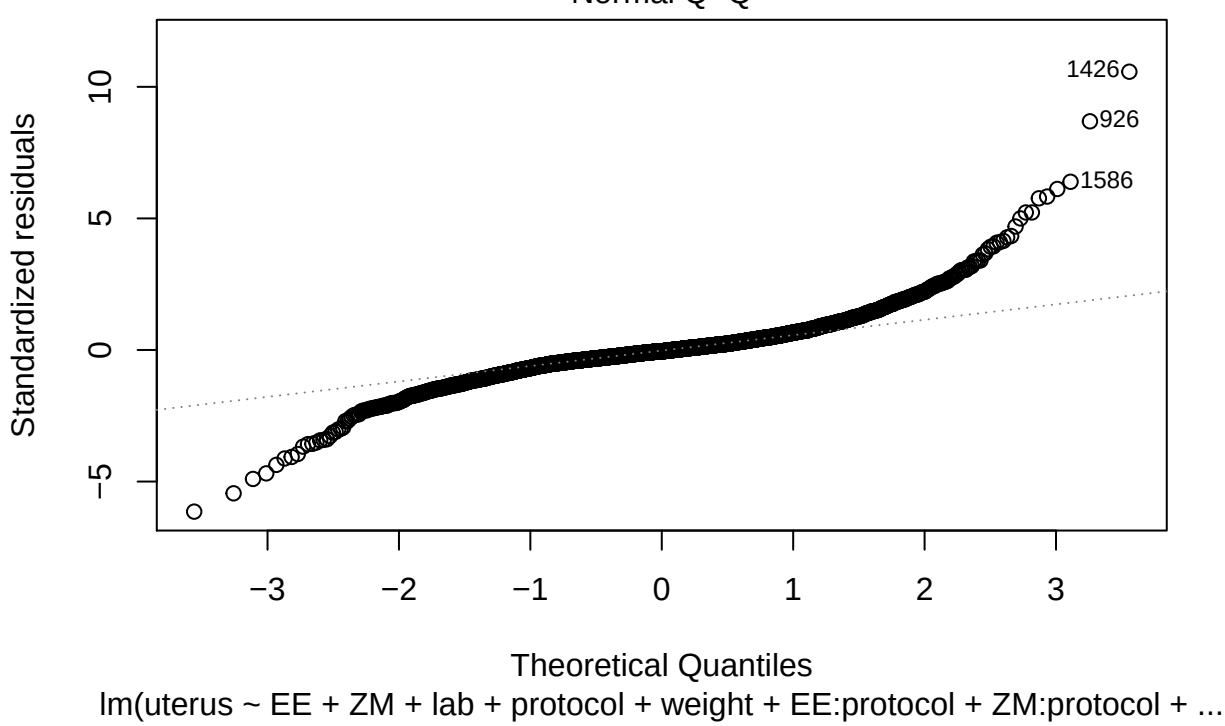
```
##without 3 interaction--simple
lm.full.fac.2=lm(data = bioassay.fac,formula = uterus~EE+ZM+lab+protocol+weight+EE:protocol+ZM:protocol
anova(lm.full.fac.2)
```

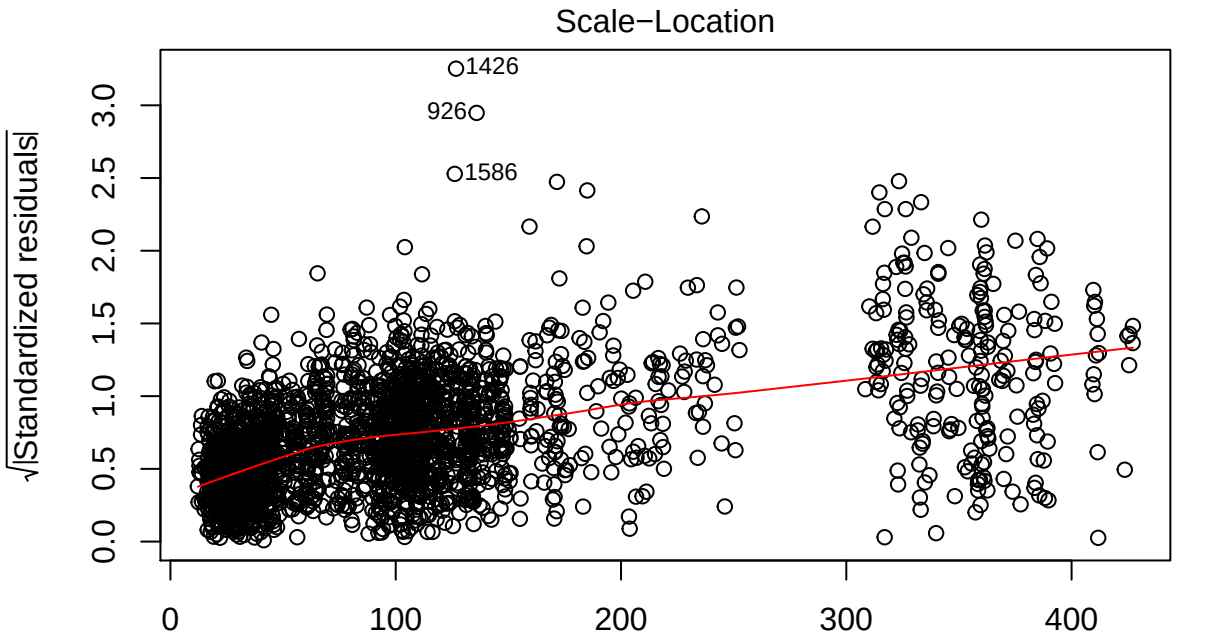
```
## Analysis of Variance Table
##
## Response: uterus
##                Df  Sum Sq Mean Sq   F value    Pr(>F)
## EE              7 6461242  923035 2314.1917 < 2.2e-16 ***
## ZM              2 2091034 1045517 2621.2742 < 2.2e-16 ***
## lab            18 2424774  134710  337.7382 < 2.2e-16 ***
## protocol        3 7416107 2472036 6197.7795 < 2.2e-16 ***
## weight          1  117187  117187  293.8051 < 2.2e-16 ***
## EE:protocol    21 2210336  105254  263.8884 < 2.2e-16 ***
## ZM:protocol     6  963956  160659  402.7981 < 2.2e-16 ***
## EE:lab        123  307910    2503    6.2763 < 2.2e-16 ***
## ZM:lab         36  105719    2937    7.3626 < 2.2e-16 ***
```

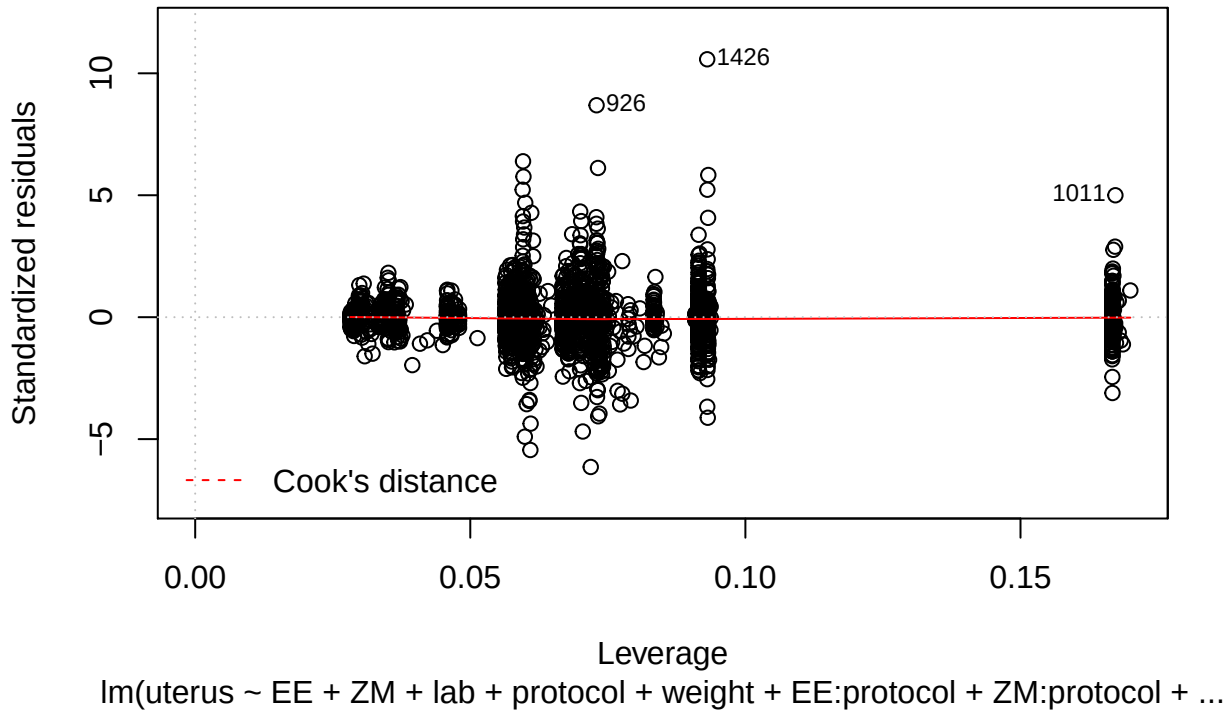
```
## Residuals   2459  980793     399
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
plot(lm.full.fac.2)
```










```
summary(lm.full.fac.2)
```

```
##
## Call:
## lm(formula = uterus ~ EE + ZM + lab + protocol + weight + EE:protocol +
##     ZM:protocol + EE:lab + ZM:lab, data = bioassay.fac)
```

```
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -118.184   -7.993   -0.927    7.132  201.127
##
## Coefficients: (3 not defined because of singularities)
##                      Estimate Std. Error t value Pr(>|t|)
## (Intercept)          17.69214    6.22835   2.841 0.004540 **
## EE0.01                1.77014    6.37095   0.278 0.781154
## EE0.03                1.03789   10.13593   0.102 0.918450
## EE0.1                 2.74322   10.13618   0.271 0.786694
## EE0.3                -1.22498   10.13589  -0.121 0.903815
## EE1                   4.91547   10.13590   0.485 0.627750
## EE3                  69.31156   10.13601   6.838 1.01e-11 ***
## EE10                 85.63718   10.13632   8.449  < 2e-16 ***
## ZM0.1               -44.26663   11.53055  -3.839 0.000127 ***
## ZM1                 -68.46723   11.53052  -5.938 3.29e-09 ***
## labBayer             13.55994    8.34848   1.624 0.104453
## labBerlin             5.12559    8.36556   0.613 0.540131
## labChungKor          16.99586    7.38751   2.301 0.021496 *
## labCitfranc          19.91352    8.33660   2.389 0.016984 *
## labCitijapa          13.44300    7.05807   1.905 0.056945 .
## labDenmark            9.58772    8.72219   1.099 0.271774
## labExxon             15.66044   10.13611   1.545 0.122471
## labHatano            11.94981    6.90834   1.730 0.083798 .
## labHuntingd          -8.13946    9.01129  -0.903 0.366481
## labInEnvTox           4.71938    6.95727   0.678 0.497622
## labKoreaPar          -2.44600    7.65622  -0.319 0.749390
## labMitsubis          11.95768    6.90938   1.731 0.083641 .
## labNihon              5.90045    6.90977   0.854 0.393228
## labPoulenc           -3.87674    8.34574  -0.465 0.642318
## labSumitomo          10.71023    7.05716   1.518 0.129233
## labTNO                5.12443    7.45549   0.687 0.491935
## labWIL                6.72808    7.39008   0.910 0.362690
## labZeneca            -4.00766    7.07787  -0.566 0.571293
## protocolB            -2.16109    2.55511  -0.846 0.397752
## protocolC            57.49670    5.53244  10.393  < 2e-16 ***
## protocolD            47.87979    6.30291   7.596 4.30e-14 ***
## weight                0.09832    0.02759   3.564 0.000372 ***
## EE0.01:protocolB      1.19030    4.41931   0.269 0.787691
## EE0.03:protocolB      3.54185    4.41931   0.801 0.422949
## EE0.1:protocolB       7.59004    4.41932   1.717 0.086020 .
## EE0.3:protocolB      35.09788    4.41931   7.942 3.00e-15 ***
## EE1:protocolB        58.21246    4.41935  13.172  < 2e-16 ***
## EE3:protocolB        34.40121    4.41934   7.784 1.02e-14 ***
## EE10:protocolB       17.57379    4.43120   3.966 7.52e-05 ***
## EE0.01:protocolC     -0.32192    5.01727  -0.064 0.948847
## EE0.03:protocolC      2.72356    5.01727   0.543 0.587291
## EE0.1:protocolC      16.89614    5.01815   3.367 0.000772 ***
## EE0.3:protocolC     100.19088    5.01797  19.966  < 2e-16 ***
## EE1:protocolC       186.27030    5.02262  37.086  < 2e-16 ***
## EE3:protocolC       169.96772    5.03205  33.777  < 2e-16 ***
## EE10:protocolC      157.50511    5.03917  31.256  < 2e-16 ***
## EE0.01:protocolD      6.45552    6.37066   1.013 0.311007
```

```
## EE0.03:protocolD      4.83542    6.42921   0.752 0.452062
## EE0.1:protocolD      31.10328    6.37787   4.877 1.15e-06 ***
## EE0.3:protocolD     140.95282    6.38021  22.092  < 2e-16 ***
## EE1:protocolD       230.45979    6.40813  35.964  < 2e-16 ***
## EE3:protocolD       208.07221    6.42688  32.375  < 2e-16 ***
## EE10:protocolD      216.86988    6.44492  33.650  < 2e-16 ***
## ZM0.1:protocolB     -61.03640    5.09942 -11.969  < 2e-16 ***
## ZM1:protocolB       -53.86498    5.09938 -10.563  < 2e-16 ***
## ZM0.1:protocolC    -158.38768    5.81546 -27.236  < 2e-16 ***
## ZM1:protocolC      -171.30052    5.79644 -29.553  < 2e-16 ***
## ZM0.1:protocolD    -183.58126    7.37935 -24.878  < 2e-16 ***
## ZM1:protocolD      -217.19102    7.37553 -29.448  < 2e-16 ***
## EE0.01:labBayer            NA         NA      NA       NA
## EE0.03:labBayer            NA         NA      NA       NA
## EE0.1:labBayer       -1.94135   14.22857  -0.136 0.891484
## EE0.3:labBayer       -1.07592   14.22866  -0.076 0.939731
## EE1:labBayer          2.43276   14.22897   0.171 0.864260
## EE3:labBayer        -28.20538   14.22855  -1.982 0.047556 *
## EE10:labBayer        13.37183   14.22881   0.940 0.347427
## EE0.01:labBerlin     -0.87184   11.84482  -0.074 0.941330
## EE0.03:labBerlin      2.90653   14.22852   0.204 0.838155
## EE0.1:labBerlin      -2.79736   14.22927  -0.197 0.844163
## EE0.3:labBerlin      15.63095   14.22866   1.099 0.272070
## EE1:labBerlin        49.48387   14.22852   3.478 0.000514 ***
## EE3:labBerlin        36.19298   14.22869   2.544 0.011030 *
## EE10:labBerlin        5.95411   14.23030   0.418 0.675683
## EE0.01:labChungKor   -4.42280    9.23342  -0.479 0.631982
## EE0.03:labChungKor    3.65423   12.54898   0.291 0.770925
## EE0.1:labChungKor    25.05789   12.54930   1.997 0.045962 *
## EE0.3:labChungKor    29.96319   12.54909   2.388 0.017030 *
## EE1:labChungKor      33.48666   12.54901   2.668 0.007670 **
## EE3:labChungKor       5.87946   12.54918   0.469 0.639459
## EE10:labChungKor    -13.13023   12.55008  -1.046 0.295559
## EE0.01:labCitfranc   -5.46736   11.84512  -0.462 0.644430
## EE0.03:labCitfranc    2.10257   14.22852   0.148 0.882535
## EE0.1:labCitfranc    -4.76553   14.22874  -0.335 0.737712
## EE0.3:labCitfranc     4.49007   14.22853   0.316 0.752357
## EE1:labCitfranc      15.39651   14.22854   1.082 0.279320
## EE3:labCitfranc     -11.45162   14.22854  -0.805 0.420993
## EE10:labCitfranc     12.32382   14.22869   0.866 0.386506
## EE0.01:labCitijapa   -3.02355    8.15367  -0.371 0.710802
## EE0.03:labCitijapa   -4.58595   11.97179  -0.383 0.701706
## EE0.1:labCitijapa    -7.09879   11.97184  -0.593 0.553264
## EE0.3:labCitijapa    -3.27505   11.97170  -0.274 0.784441
## EE1:labCitijapa      35.06124   11.97183   2.929 0.003436 **
## EE3:labCitijapa       5.05010   11.97169   0.422 0.673181
## EE10:labCitijapa     10.56809   11.97209   0.883 0.377470
## EE0.01:labDenmark    -2.11565   11.81841  -0.179 0.857942
## EE0.03:labDenmark    -3.87161   14.89923  -0.260 0.794999
## EE0.1:labDenmark     -5.42759   14.89906  -0.364 0.715672
## EE0.3:labDenmark     27.69075   14.89904   1.859 0.063209 .
## EE1:labDenmark       40.24816   14.89938   2.701 0.006954 **
## EE3:labDenmark        8.87246   14.89913   0.596 0.551563
## EE10:labDenmark      -2.30835   14.90258  -0.155 0.876916
```

```
## EE0.01:labExxon       5.24081   13.17346   0.398 0.690789
## EE0.03:labExxon       0.66382   15.35219   0.043 0.965514
## EE0.1:labExxon       -0.36748   15.35261  -0.024 0.980906
## EE0.3:labExxon        9.46326   15.35218   0.616 0.537680
## EE1:labExxon         11.90863   15.35219   0.776 0.438003
## EE3:labExxon          5.48799   15.35267   0.357 0.720777
## EE10:labExxon        14.68112   15.35245   0.956 0.339029
## EE0.01:labHatano     -3.13917    7.76232  -0.404 0.685945
## EE0.03:labHatano     -3.64550   11.71067  -0.311 0.755601
## EE0.1:labHatano       2.00726   11.70853   0.171 0.863895
## EE0.3:labHatano       2.92416   11.70853   0.250 0.802805
## EE1:labHatano        25.97766   11.70862   2.219 0.026600 *
## EE3:labHatano        14.26924   11.70864   1.219 0.223077
## EE10:labHatano       23.85526   11.73860   2.032 0.042240 *
## EE0.01:labHuntingd    7.87841   11.89753   0.662 0.507913
## EE0.03:labHuntingd   -5.14082   15.08736  -0.341 0.733331
## EE0.1:labHuntingd   -32.88155   15.08992  -2.179 0.029423 *
## EE0.3:labHuntingd   -91.01782   15.08977  -6.032 1.87e-09 ***
## EE1:labHuntingd    -151.30544   15.08721 -10.029  < 2e-16 ***
## EE3:labHuntingd    -101.12490   15.09356  -6.700 2.58e-11 ***
## EE10:labHuntingd     -5.29492   15.08902  -0.351 0.725684
## EE0.01:labInEnvTox   -5.50629    7.76187  -0.709 0.478142
## EE0.03:labInEnvTox   -2.50094   11.71053  -0.214 0.830905
## EE0.1:labInEnvTox    -0.34155   11.70970  -0.029 0.976733
## EE0.3:labInEnvTox     8.10498   11.70854   0.692 0.488859
## EE1:labInEnvTox      35.79794   11.70873   3.057 0.002257 **
## EE3:labInEnvTox       5.81048   11.70908   0.496 0.619772
## EE10:labInEnvTox     -5.14531   11.70920  -0.439 0.660393
## EE0.01:labKoreaPar   -4.56482    9.21597  -0.495 0.620421
## EE0.03:labKoreaPar   -9.04788   13.00478  -0.696 0.486661
## EE0.1:labKoreaPar   -13.56872   13.00520  -1.043 0.296898
## EE0.3:labKoreaPar    -8.21180   13.00478  -0.631 0.527808
## EE1:labKoreaPar      32.49925   13.00499   2.499 0.012520 *
## EE3:labKoreaPar      -5.68840   13.00542  -0.437 0.661869
## EE10:labKoreaPar     10.68224   13.00612   0.821 0.411541
## EE0.01:labMitsubis   -6.96729    7.76209  -0.898 0.369484
## EE0.03:labMitsubis   -3.65380   11.72661  -0.312 0.755384
## EE0.1:labMitsubis    -2.43796   11.70853  -0.208 0.835074
## EE0.3:labMitsubis    10.14575   11.70869   0.867 0.386293
## EE1:labMitsubis      23.99060   11.70930   2.049 0.040583 *
## EE3:labMitsubis      12.01364   11.70891   1.026 0.304981
## EE10:labMitsubis     10.19397   11.70866   0.871 0.384039
## EE0.01:labNihon       1.13423    7.76226   0.146 0.883838
## EE0.03:labNihon      -6.85430   11.71053  -0.585 0.558393
## EE0.1:labNihon      -11.04751   11.70863  -0.944 0.345499
## EE0.3:labNihon       -4.31926   11.70878  -0.369 0.712241
## EE1:labNihon         22.31761   11.70856   1.906 0.056755 .
## EE3:labNihon         16.69601   11.70854   1.426 0.154004
## EE10:labNihon        16.71569   11.70878   1.428 0.153528
## EE0.01:labPoulenc    -2.67678   11.84484  -0.226 0.821230
## EE0.03:labPoulenc    -1.08198   14.22852  -0.076 0.939391
## EE0.1:labPoulenc     -2.12507   14.22865  -0.149 0.881289
## EE0.3:labPoulenc     15.62998   14.22861   1.098 0.272098
## EE1:labPoulenc       31.54972   14.22852   2.217 0.026690 *
```

```
## EE3:labPoulenc      -17.87466   14.22859  -1.256 0.209145
## EE10:labPoulenc     -25.63187   14.22878  -1.801 0.071761 .
## EE0.01:labSumitomo   -0.77958    8.15347  -0.096 0.923836
## EE0.03:labSumitomo   -1.56877   11.97178  -0.131 0.895755
## EE0.1:labSumitomo    -6.93544   11.97186  -0.579 0.562432
## EE0.3:labSumitomo    11.70930   11.97174   0.978 0.328132
## EE1:labSumitomo      42.03130   11.97170   3.511 0.000455 ***
## EE3:labSumitomo      24.45385   11.97179   2.043 0.041196 *
## EE10:labSumitomo     35.65961   11.97207   2.979 0.002924 **
## EE0.01:labTNO         0.14093    9.27190   0.015 0.987874
## EE0.03:labTNO        -0.93509   12.58347  -0.074 0.940769
## EE0.1:labTNO         -5.59135   12.58363  -0.444 0.656840
## EE0.3:labTNO          2.75133   12.58352   0.219 0.826945
## EE1:labTNO           32.73828   12.58354   2.602 0.009333 **
## EE3:labTNO           10.96012   12.58351   0.871 0.383844
## EE10:labTNO          20.51929   12.58495   1.630 0.103132
## EE0.01:labWIL         2.38794    9.23282   0.259 0.795938
## EE0.03:labWIL        -0.99647   12.54895  -0.079 0.936716
## EE0.1:labWIL          1.57193   12.54958   0.125 0.900330
## EE0.3:labWIL         -0.58510   12.54903  -0.047 0.962816
## EE1:labWIL           13.04750   12.54905   1.040 0.298572
## EE3:labWIL          -12.00282   12.54936  -0.956 0.338940
## EE10:labWIL           4.31681   12.55064   0.344 0.730913
## EE0.01:labZeneca           NA         NA      NA       NA
## EE0.03:labZeneca      0.24516   11.97179   0.020 0.983664
## EE0.1:labZeneca       0.17826   11.97205   0.015 0.988121
## EE0.3:labZeneca      13.26839   11.97171   1.108 0.267836
## EE1:labZeneca        21.78971   11.97171   1.820 0.068865 .
## EE3:labZeneca       -23.55358   11.97193  -1.967 0.049249 *
## EE10:labZeneca      -16.89659   11.97261  -1.411 0.158292
## ZM0.1:labBayer       50.88153   16.30681   3.120 0.001828 **
## ZM1:labBayer         32.78089   16.30662   2.010 0.044510 *
## ZM0.1:labBerlin      27.72458   16.30661   1.700 0.089219 .
## ZM1:labBerlin        -2.22047   16.30662  -0.136 0.891698
## ZM0.1:labChungKor    57.68753   14.35039   4.020 6.00e-05 ***
## ZM1:labChungKor      33.01889   14.35042   2.301 0.021481 *
## ZM0.1:labCitfranc    30.20924   16.30667   1.853 0.064064 .
## ZM1:labCitfranc      12.73812   16.30663   0.781 0.434782
## ZM0.1:labCitijapa    31.79891   13.67781   2.325 0.020161 *
## ZM1:labCitijapa       7.74008   13.67687   0.566 0.571497
## ZM0.1:labDenmark     23.13636   17.08541   1.354 0.175810
## ZM1:labDenmark        8.21386   17.08538   0.481 0.630734
## ZM0.1:labExxon       23.05588   16.30689   1.414 0.157526
## ZM1:labExxon         -5.21047   16.30673  -0.320 0.749353
## ZM0.1:labHatano       8.99883   13.39223   0.672 0.501682
## ZM1:labHatano         0.30580   13.36968   0.023 0.981754
## ZM0.1:labHuntingd    72.42672   17.31097   4.184 2.97e-05 ***
## ZM1:labHuntingd     114.05654   17.30486   6.591 5.33e-11 ***
## ZM0.1:labInEnvTox    23.32655   13.36992   1.745 0.081161 .
## ZM1:labInEnvTox      10.23879   13.36964   0.766 0.443854
## ZM0.1:labKoreaPar    16.15753   14.88321   1.086 0.277753
## ZM1:labKoreaPar      40.03131   14.88130   2.690 0.007193 **
## ZM0.1:labMitsubis    13.24888   13.36992   0.991 0.321809
## ZM1:labMitsubis      -5.99038   13.36964  -0.448 0.654151
```

```
## ZM0.1:labNihon       12.59112   13.36996   0.942 0.346415
## ZM1:labNihon         -4.32055   13.36965  -0.323 0.746601
## ZM0.1:labPoulenc     49.01203   16.30661   3.006 0.002677 **
## ZM1:labPoulenc       29.60331   16.30663   1.815 0.069582 .
## ZM0.1:labSumitomo    30.87219   13.67789   2.257 0.024090 *
## ZM1:labSumitomo      -7.69915   13.67692  -0.563 0.573533
## ZM0.1:labTNO         77.47526   14.35033   5.399 7.35e-08 ***
## ZM1:labTNO           22.63849   14.35027   1.578 0.114794
## ZM0.1:labWIL         38.18457   14.35029   2.661 0.007844 **
## ZM1:labWIL           29.80515   14.35027   2.077 0.037908 *
## ZM0.1:labZeneca      77.94834   13.67822   5.699 1.35e-08 ***
## ZM1:labZeneca        44.78820   13.67691   3.275 0.001072 **
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## Residual standard error: 19.97 on 2459 degrees of freedom
##   (4 observations deleted due to missingness)
## Multiple R-squared:  0.9575, Adjusted R-squared:  0.9538
## F-statistic: 255.3 on 217 and 2459 DF,  p-value: < 2.2e-16
```

```
#Adjusted R-squared:  0.9538

lm.full.q=lm(data = bioassay,formula = uterus~poly(EE,2)+poly(ZM,2)+lab+protocol+weight+EE:protocol+ZM:p
summary(lm.full.q) #0.8286#p=69
```

```
##
## Call:
## lm(formula = uterus ~ poly(EE, 2) + poly(ZM, 2) + lab + protocol +
##     weight + EE:protocol + ZM:protocol + EE:lab + ZM:lab, data = bioassay)
##
## Residuals:
##      Min       1Q   Median       3Q      Max
## -129.731  -19.212   -2.043   13.294  227.887
##
## Coefficients: (2 not defined because of singularities)
##                   Estimate Std. Error t value Pr(>|t|)
## (Intercept)     7.203e+01  6.080e+00  11.846  < 2e-16 ***
## poly(EE, 2)1    4.374e+03  3.006e+02  14.551  < 2e-16 ***
## poly(EE, 2)2   -2.297e+03  5.639e+01 -40.729  < 2e-16 ***
## poly(ZM, 2)1   -3.580e+03  2.972e+02 -12.046  < 2e-16 ***
## poly(ZM, 2)2    1.284e+03  4.797e+01  26.770  < 2e-16 ***
## labBayer        3.473e+00  9.278e+00   0.374  0.70818
## labBerlin       1.442e+01  8.636e+00   1.669  0.09515 .
## labChungKor     3.309e+01  7.627e+00   4.338 1.49e-05 ***
## labCitfranc     2.324e+01  8.543e+00   2.721  0.00656 **
## labCitijapa     1.815e+01  7.300e+00   2.486  0.01298 *
## labDenmark      1.916e+01  8.929e+00   2.146  0.03197 *
## labExxon        2.156e+01  8.843e+00   2.438  0.01484 *
## labHatano       1.957e+01  7.152e+00   2.736  0.00625 **
## labHuntingd    -2.560e+01  9.749e+00  -2.626  0.00868 **
## labInEnvTox     4.964e+00  7.304e+00   0.680  0.49674
## labKoreaPar    -6.208e+00  7.905e+00  -0.785  0.43231
## labMitsubis     2.055e+01  7.154e+00   2.872  0.00411 **
## labNihon        8.404e+00  7.158e+00   1.174  0.24053
## labPoulenc      1.397e+00  8.576e+00   0.163  0.87057
```

```
## labSumitomo     2.092e+01  7.296e+00   2.867  0.00418 **
## labTNO          1.129e+01  7.675e+00   1.470  0.14156
## labWIL          8.616e+00  7.629e+00   1.129  0.25888
## labZeneca       1.237e+01  7.370e+00   1.679  0.09336 .
## protocolB       1.064e+01  2.556e+00   4.163 3.25e-05 ***
## protocolC       1.534e+02  9.252e+00  16.583  < 2e-16 ***
## protocolD       1.609e+02  9.992e+00  16.105  < 2e-16 ***
## weight         -2.221e-01  5.207e-02  -4.266 2.06e-05 ***
## protocolA:EE   -1.770e+01  1.079e+00 -16.406  < 2e-16 ***
## protocolB:EE   -1.716e+01  1.074e+00 -15.983  < 2e-16 ***
## protocolC:EE   -4.325e+00  1.085e+00  -3.986 6.91e-05 ***
## protocolD:EE           NA         NA      NA       NA
## protocolA:ZM    1.267e+02  1.066e+01  11.878  < 2e-16 ***
## protocolB:ZM    8.903e+01  1.058e+01   8.413  < 2e-16 ***
## protocolC:ZM    3.592e+01  1.083e+01   3.318  0.00092 ***
## protocolD:ZM           NA         NA      NA       NA
## labBayer:EE     1.723e+00  2.440e+00   0.706  0.48005
## labBerlin:EE    1.039e+00  2.388e+00   0.435  0.66355
## labChungKor:EE -1.883e+00  2.109e+00  -0.893  0.37201
## labCitfranc:EE  8.594e-01  2.388e+00   0.360  0.71899
## labCitijapa:EE  1.205e+00  2.013e+00   0.599  0.54931
## labDenmark:EE  -5.723e-01  2.500e+00  -0.229  0.81897
## labExxon:EE     1.349e+00  2.411e+00   0.560  0.57584
## labHatano:EE    2.290e+00  1.975e+00   1.160  0.24618
## labHuntingd:EE  7.032e-01  2.531e+00   0.278  0.78119
## labInEnvTox:EE -5.644e-01  1.969e+00  -0.287  0.77438
## labKoreaPar:EE  1.143e+00  2.185e+00   0.523  0.60082
## labMitsubis:EE  8.644e-01  1.969e+00   0.439  0.66067
## labNihon:EE     2.087e+00  1.969e+00   1.060  0.28930
## labPoulenc:EE  -3.000e+00  2.388e+00  -1.256  0.20920
## labSumitomo:EE  3.626e+00  2.013e+00   1.802  0.07167 .
## labTNO:EE       2.721e+00  2.110e+00   1.290  0.19732
## labWIL:EE       1.130e-01  2.109e+00   0.054  0.95728
## labZeneca:EE   -1.940e+00  2.013e+00  -0.964  0.33505
## labBayer:ZM     6.401e+00  2.378e+01   0.269  0.78783
## labBerlin:ZM    1.874e+01  2.368e+01   0.791  0.42886
## labChungKor:ZM  3.036e+01  2.085e+01   1.456  0.14555
## labCitfranc:ZM -3.624e+00  2.368e+01  -0.153  0.87838
## labCitijapa:ZM  5.395e+00  1.988e+01   0.271  0.78611
## labDenmark:ZM   8.949e+00  2.481e+01   0.361  0.71830
## labExxon:ZM    -7.844e+00  2.372e+01  -0.331  0.74092
## labHatano:ZM    2.498e+00  1.944e+01   0.129  0.89774
## labHuntingd:ZM  5.499e+01  2.513e+01   2.189  0.02871 *
## labInEnvTox:ZM  1.146e+01  1.943e+01   0.589  0.55561
## labKoreaPar:ZM  3.035e+01  2.162e+01   1.404  0.16051
## labMitsubis:ZM -2.852e+00  1.943e+01  -0.147  0.88334
## labNihon:ZM     3.984e+00  1.943e+01   0.205  0.83757
## labPoulenc:ZM   1.357e+01  2.368e+01   0.573  0.56651
## labSumitomo:ZM -2.127e+00  1.988e+01  -0.107  0.91481
## labTNO:ZM       2.081e+01  2.085e+01   0.998  0.31839
## labWIL:ZM       1.478e+01  2.085e+01   0.709  0.47855
## labZeneca:ZM    2.309e+01  1.988e+01   1.162  0.24546
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
##
## Residual standard error: 38.44 on 2608 degrees of freedom
##   (4 observations deleted due to missingness)
## Multiple R-squared:  0.833,  Adjusted R-squared:  0.8286
## F-statistic: 191.3 on 68 and 2608 DF,  p-value: < 2.2e-16
```

4.Results

5.Conclusions

Part I: OLS/MLE