

Study Report:

Effect of supplementation of MICRO – COMPLETE – a product from PROBYN INTERNATIONAL INC., USA on performance and gut health in layer chickens

Introduction

In layer poultry farming, pullet period is critical to prepare for laying and also during the laying period especially at peak, layers might be very sensitive to diseases. To reduce this risk, chickens were often supplied antibiotic in feed or drinking water for some certain times or even more. It is the big public health concern due to the problem of residues and antibiotic resistance. Probiotic and prebiotic are the well-known alternatives for antibiotic replacement of this purpose. However, many commercial products are available without knowing their clear efficacy. A new product in Vietnam market which is specific for chicken, **MICRO – COMPLETE** – a product from **PROBYN** company - containing digestive enzymes, lactic acid bacteria and *Saccharomyces cerevisiae*) is a potential solution to improve gut health and then performance in layer chicken from pullet to laying period. This study was carried out to evaluate the effect of the product.

Materials and Methods

Study design

The study was conducted from January 2019 to June 2019 at Anh-Duong farm, Ben Cat District, Binh Duong Province. This is a commercial large-scale layer farm. On the farm, 2 types of chicken were the target of the study: pullet from 8 to 14 weeks of age and laying hen from 25 to 31 weeks of age. These chickens were reared in difference houses of 21,000 chickens. For each type of chicken, there were 2 groups: control group and treatment group (MICRO – COMPLETE supplementation with the dose of 500g / ton of feed). Other factors such as husbandry management, vaccination program was followed to farm's schedule, and were the same for both two groups.

Data collection

During study, performance data would be collected from week-age 15 to 19 in pullets and 25-29 in layer. Data maybe collected daily or weekly with a number of chickens in the groups on the base of type of parameters. The parameters to be collected are described below

- (i) **Production indicators** such as body weight, feed consumption in pullet, and egg production in layers. Because data were collected in a representative individual/ group/ house, the number of observations might be different.
- (ii) **Mortality**: number of deaths per 1000 chicks could be calculated daily or aggregated into week

- (iii) **Daily fecal scores** were performed. For each group, at least 50 fecal clumps were evaluated in the scale from 0-3 (0: small and compact dropping with white cap, no sign of wetness; 1: increase size, bulky, oily, moist; 2: watery, undigested feed, loss of firmness and sloughed off mucosa; 3: No consistency at all (no shape), watery, mucus, undigested feed and foamy)
- (iv) **Intestinal microflora** was evaluated by counting the number of bacteria in a gram of feces (*Clostridium perfringens*, *Lactobacillus*, *E.coli*, and *Bacillus*). There were 2 points of time for sampling: before the study and after the supplement for 4 weeks. For each point of sampling, 5 chickens from each group were sent to the Vet hospital (Nong Lam University) for fecal sampling and autopsy. Fecal samples were sent to the microbiological lab of the hospital to count for those bacteria following the conventional method of plate count of the lab.
- (v) **Length of villi and depth of crypt** in the duodenum: The chicken sent to the vet hospital were also performed autopsy and intestine were collected to send to the lab to make a special staining and measure the villus length and crypt depth to assess gut health of study chickens.

Data analysis

Data were managed in MS Excel 2013 then export to Stata 14 (StataCorp. 2015. College Station, TX: StataCorp LP) for statistical analysis. Feed consumption, body weight, villus lengths, and crypt depths between 2 groups in each type of chicken were compared using ANOVA. Egg production and mortality data seem to be the count number over a quantity, Poisson regression were used to assess the effect of treatment comparing to control group via coefficient values. These values can be transport to natural exponential forms to understand how many times of increase/ decrease of the treatment group to the control group in terms of the interested parameters. For fecal scores which are ordered data, the comparison between 2 groups would be performed by non-parametric Kruskal-Wallis test.

Results

- ***Growth indicators of pullets***

Although pullets were not considered much on the growth rate, their weight should be controlled very well, not too skinny and not too fat, and high uniformity. Table 1 and Figure 1 show weekly body weights of pullets in the study respectively. The weights in treatment group seemed to vary lower than in the control group with **smaller value of SD** (standard deviation). There is no difference between 2 groups during the later weeks. Table 2 and Figure 2 show the feed consumption of the 2 groups. It can be seen that the treatment group can have more feed consumption, however, there is not significant difference. The number of data is small due to the pool data - individual bird or pens could not be achieved. From

this point of finding, the supplement prepared a good start for pullets to go to the laying stage.

Table 1. Weekly weights (gram) of pullets in the study

Week age	Number of weighted birds	Control group		Treatment group		p value
		Mean	SD	Mean	SD	
15	80	943.3	128.2	980.8	109.2	0.0482
16	80	1085.3	105.9	1067.4	116.0	0.3102
17	80	1160.3	86.6	1128.4	105.4	0.0382
18	80	1179.5	109.8	1185.0	108.0	0.7504
19	80	1254.9	130.1	1247.8	109.0	0.7077

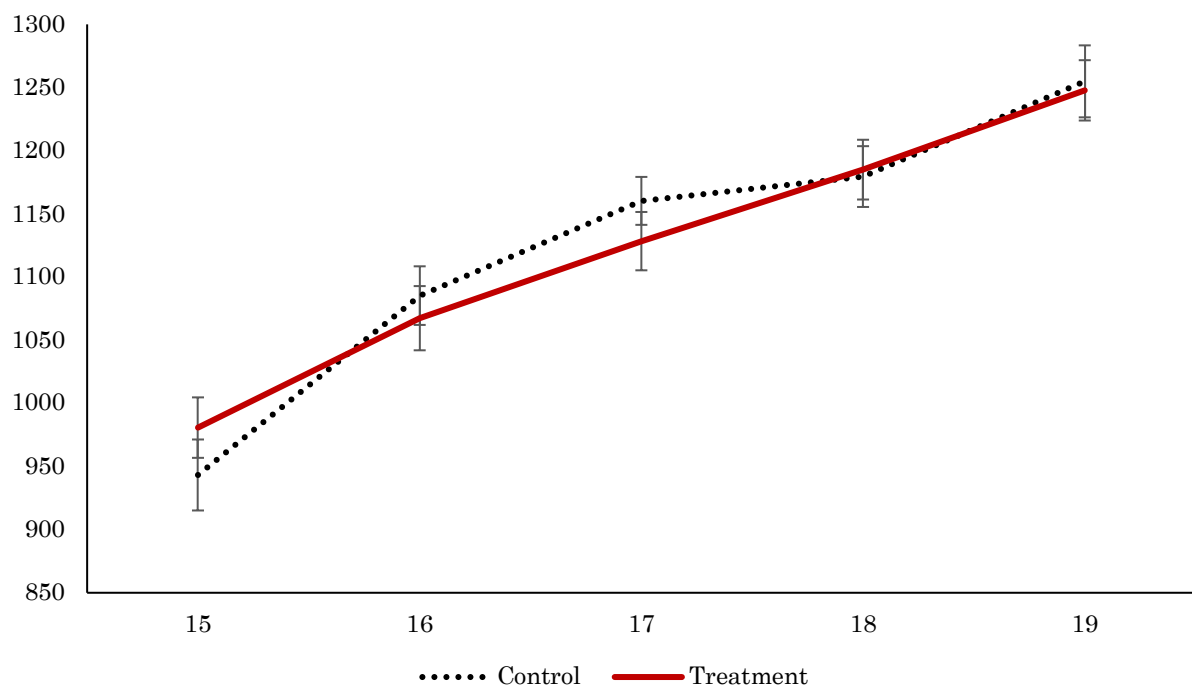


Figure 1. Weekly body weights (gram) with their 95% of confident interval of pullets in two groups

Table 2. Average feed consumption (gram/bird/day) of pullets by weeks in the study

Week age	Number of data	Control group		Treatment group		p value
		Mean	SD	Mean	SD	
15	12	62.0	6.1	58.4	2.8	0.0090
16	14	69.9	4.7	66.4	3.2	0.0120
17	14	65.4	5.0	62.0	5.8	0.0230
18	14	55.0	10.6	63.8	7.6	0.0120
19	2	60.2	5.8	68.8	11.0	0.1700

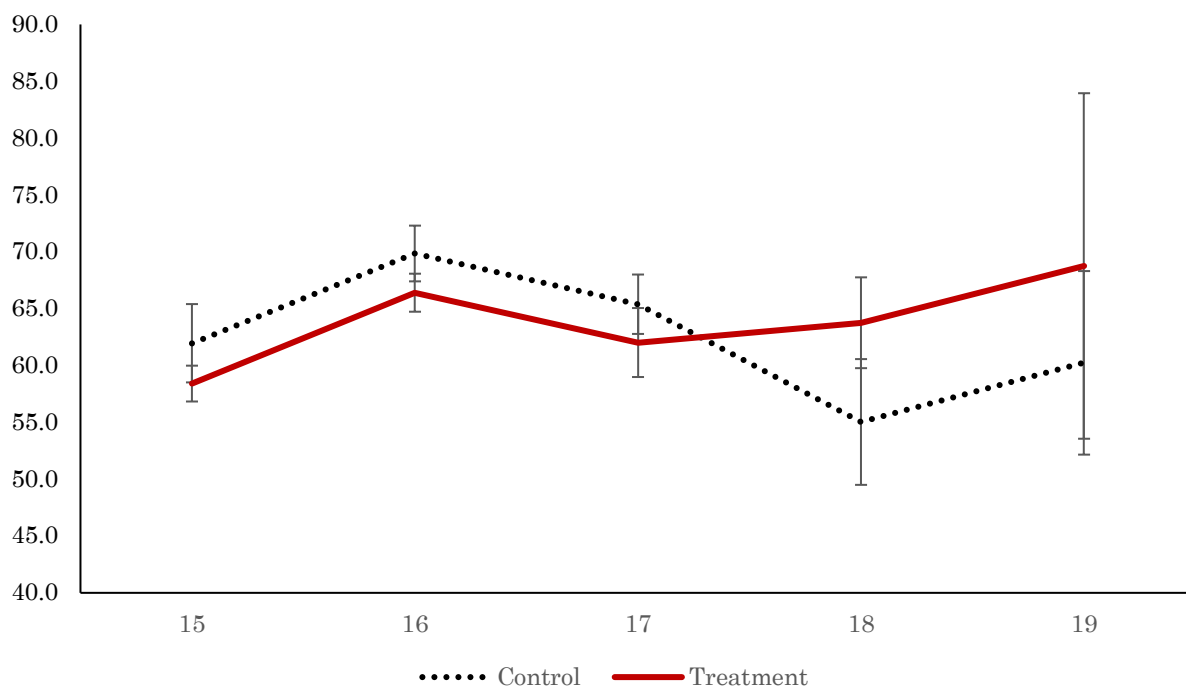


Figure 2. Average feed consumption (gram/bird/day) of pullets by week age in the study

- ***Egg production in layers***

The number of eggs from layer per day could be calculated from the aggregated report by week to make the number more meaningful. Table 3 and Figure 3 show this result. Poisson regression revealed that the amount of egg/bird/day in treatment group would be 1.0036 time of that in control group statistically significantly ($p= 0.016$). It can be simply understood that in the case we have the same number of layer (i.e., 1000) and the control group can produce 1000 egg per day, then the treatment group with the same number of layers would produce 1008.4 eggs. That means using MICRO – COMPLETE can increase egg production in layer.

Table 3. Average egg production (egg/bird/day) of layer by weeks in the study

Week age	Number of data	Control group		Treatment group	
		Mean	SD	Mean	SD
24	2	0.847	0.010	0.862	0.006
25	7	0.900	0.018	0.897	0.012
26	7	0.942	0.039	0.922	0.001
27	7	0.936	0.000	0.936	0.006
28	7	0.934	0.003	0.944	0.001
29	7	0.930	0.010	0.946	0.001
30	5	0.927	0.000	0.945	0.004

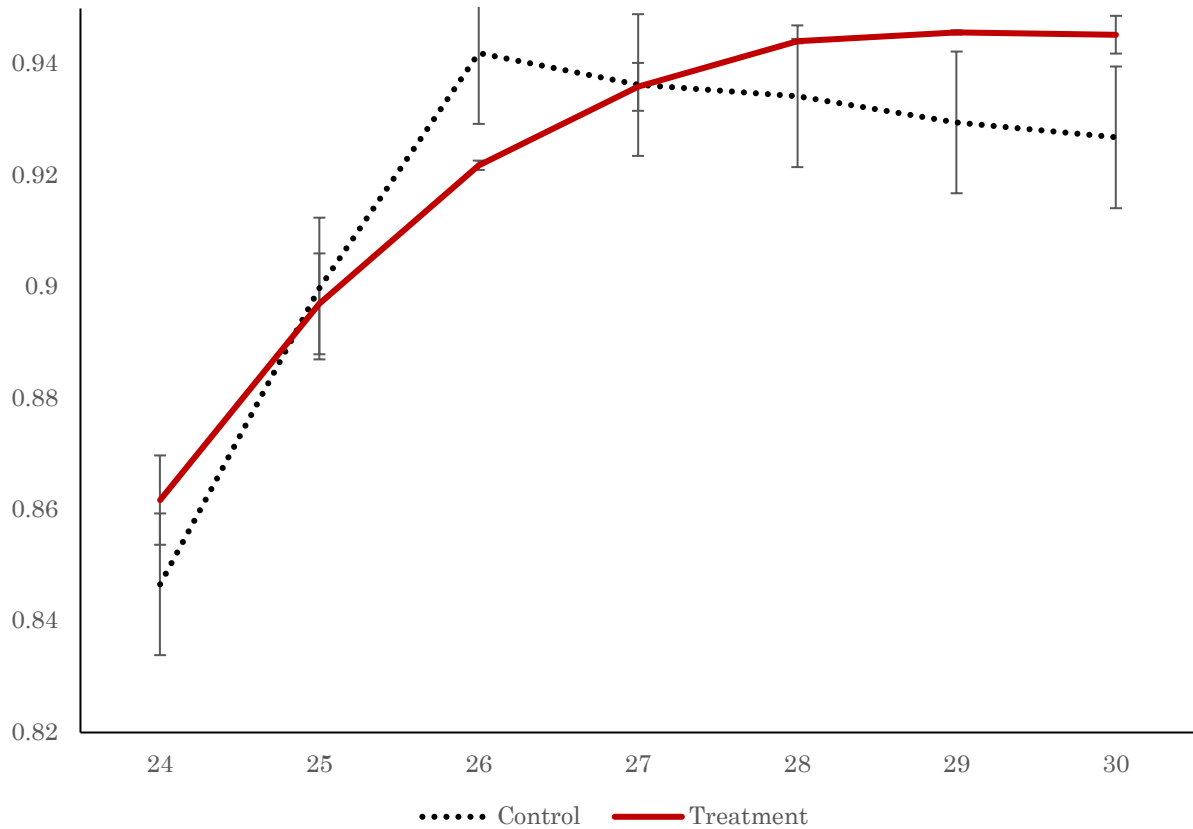


Figure 3. Average egg production (egg/bird/day) of layer by weeks in the study

- ***Mortality***

Number of deaths per 1000 bird per day can be seen in Table 4 and Figure 4 for pullets and Table 5 and Figure 5 for layer. It can be seen that mortality in treatment group might be slightly higher than the control group, especially for layers. In layer, the mortality in treatment group is 1.2 times more than that of control group non-statistically significant ($p=0.051$).

Table 4. Average number of deaths per 1000 pullets per day by week in the study

Week age	Number of data	Control group		Treatment group	
		Mean	SD	Mean	SD
15	12	1.196	2.649	0.515	0.343
16	14	0.440	0.349	0.257	0.207
17	14	0.200	0.201	0.207	0.191
18	14	0.166	0.159	0.207	0.236
19	2	0.241	0.341	0.353	0.277
p-value		0.1480			

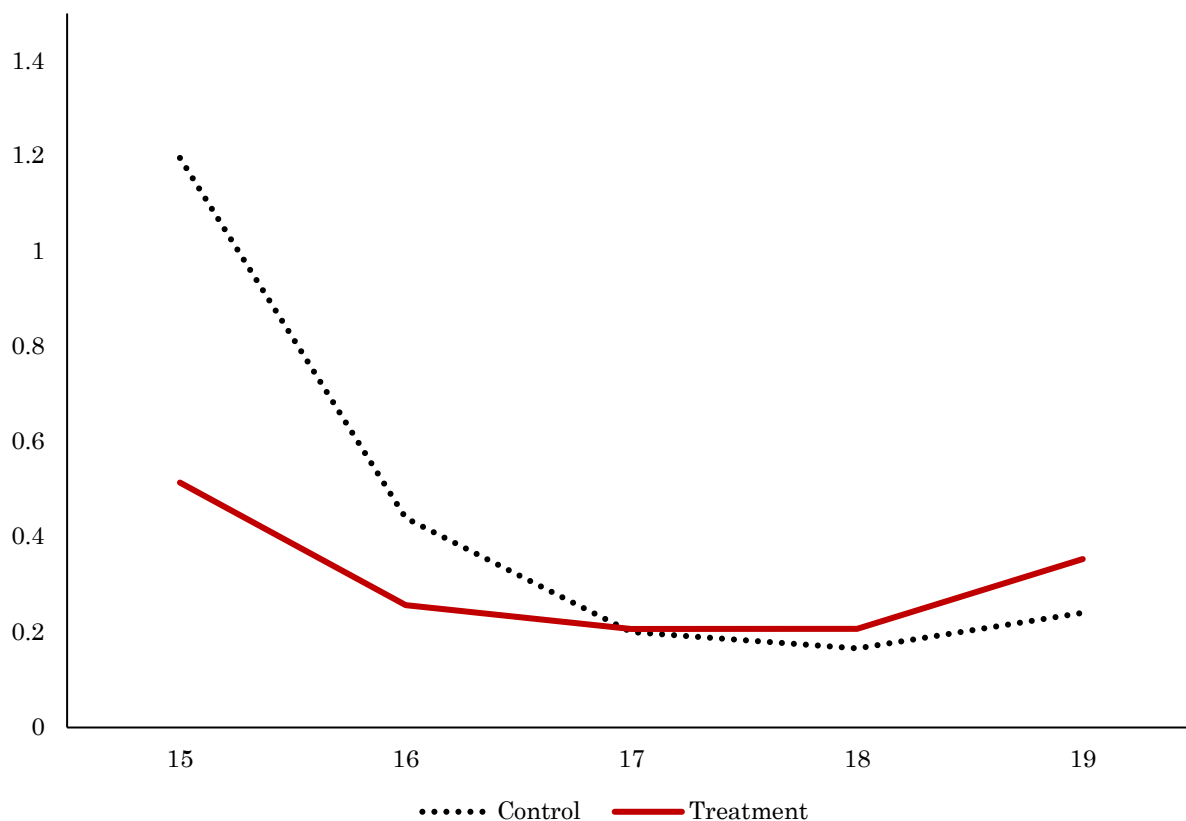


Figure 4. Average number of deaths per 1000 pullets per day by week in the study

Table 5. Average number of deaths per 1000 layers per day by week in the study

Week age	Number of data	Control group		Treatment group	
		Mean	SD	Mean	SD
24	2	0.400	0.000	0.222	0.000
25	7	0.121	0.102	0.235	0.076
26	7	0.178	0.057	0.216	0.048
27	7	0.166	0.056	0.191	0.071
28	7	0.223	0.082	0.230	0.087
29	7	0.249	0.051	0.243	0.051
30	5	0.125	0.086	0.296	0.068
p-value		0.051			

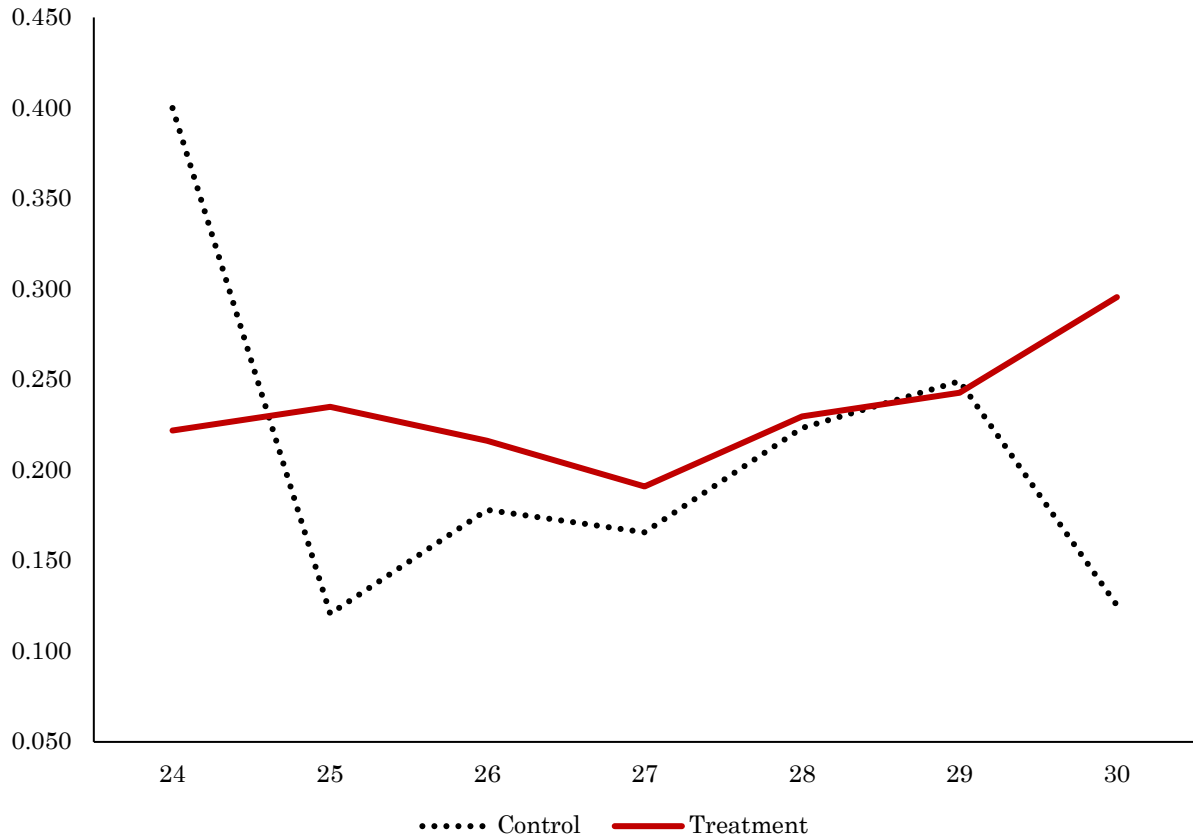


Figure 5. Average number of deaths per 1000 layers per day by week in the study

- ***Fecal score***

Feces from both groups were evaluated and classified into scores. Table 6 and Figure 6 show the distribution of these scores. Statistical analysis found that there was significant difference from median score of feces, i.e., the control group had higher score than the score of treatment groups in both pullet and layer. Table 7, Figures 7 and 8 show the mean scores of these groups. In general, using MICRO – COMPLETE can lead to improve fecal status of both layers, pullets.

Table 6. Distribution of scores of examined feces from pullets and layers in the study

Score	Pullets			Layers		
	Control	Treatment	Total	Control	Treatment	Total
0	0	1	1	49	194	243
1	1,729	1,824	3,553	521	613	1,134
2	465	379	844	433	283	716
3	46	36	82	117	30	147
Total	2,240	2,240	4,480	1,120	1,120	2,240
p- value	0.012			0.0001		



Figure 6. Distribution of scores of examined feces from pullets and layers in the study

Table 7. Distribution of scores of examined feces from pullets and layers in the study

		Pullet						Layer			
		Control		Treatment				Control		Treatment	
<i>wk.</i>	<i>n</i>	Mean	SD	Mean	SD	<i>wk.</i>	<i>n</i>	Mean	SD	Mean	SD
15	480	1.2	0.5	1.2	0.5	24	80	1.5	0.7	1.2	0.7
16	560	1.3	0.5	1.3	0.5	25	280	1.6	0.7	1.5	0.6
17	560	1.2	0.4	1.2	0.4	26	280	1.6	0.8	1.2	0.7
18	560	1.2	0.4	1.2	0.4	27	280	1.4	0.7	1.0	0.7
19	80	1.4	0.6	1.2	0.4	28	200	1.7	0.8	0.7	0.8

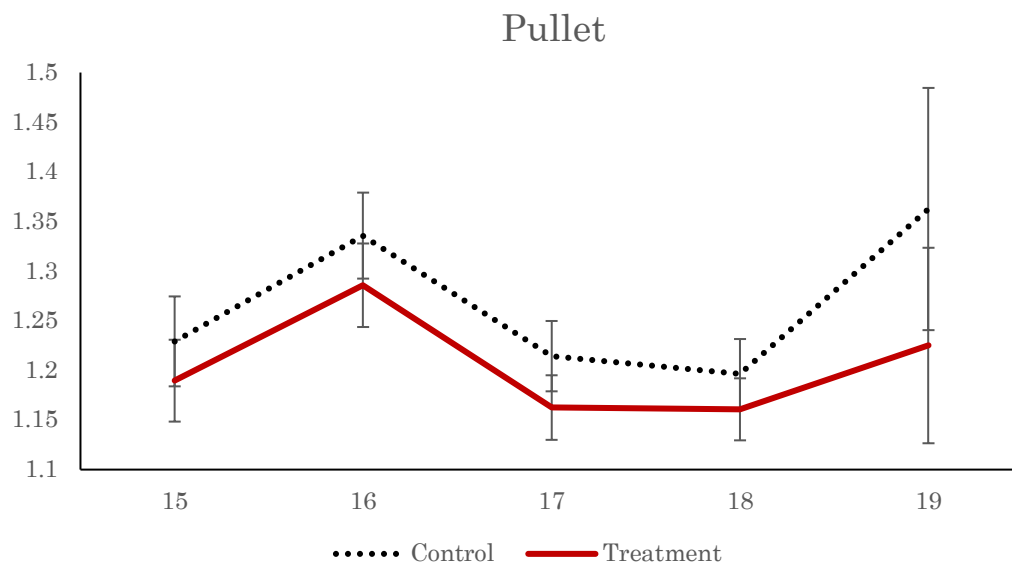


Figure 7. Mean scores with 95% confident interval of examined feces from pullets in the study

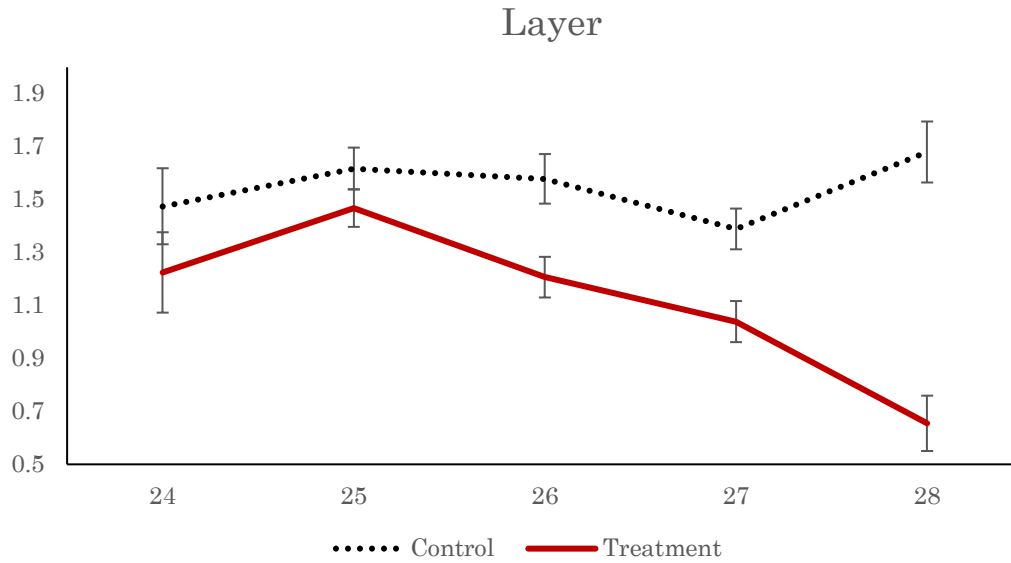


Figure 8. Mean scores with 95% confident interval of examined feces from layers in the study

- ***Intestinal bacterial enumeration***

No *Clostridium* was detected, so no statistics was carried out. Due to the balanced number of pullets (5 birds) and layers (5 birds) in both control and experimental groups, statistics was only done on control and treatment groups before and after PROBYN supplementation. Factors of breeding and chicken type were the same and then were not considered for analysis.

Table 8. Number of bacteria (log scale) in a gram of feces in 2 groups before product supplement and 4 weeks after supplement for each type of chicken

			N	<i>Lactobacillus</i>		<i>E. coli</i>		<i>Bacillus</i>	
				log	SE	log	SE	log	SE
Before	Pullet	Control	5	12.89	1.71	1.35	0.55	5.58	1.44
		Treatment	5	15.23	1.52	5.44	1.13	7.43	0.46
	Layer	Control	5	12.24	1.50	1.39	0.91	5.51	0.28
		Treatment	5	11.77	1.53	3.80	1.44	3.49	1.45
	Both	Control	10	12.57	1.08	1.37	0.43	5.54	0.69
		Treatment	10	13.50	1.17	4.62	1.46	5.46	0.97
p-value				0.5637		0.0056		0.9438	
After	Pullet	Control	5	13.16	0.70	0.38	0.23	6.83	1.78
		Treatment	5	12.02	0.62	0.79	0.76	4.14	1.69
	Layer	Control	5	13.92	0.49	3.43	0.87	5.84	2.46
		Treatment	5	13.49	0.60	3.11	0.92	7.45	1.90
	Both	Control	10	13.54	0.42	1.91	0.66	6.34	1.44
		Treatment	10	12.75	0.48	1.95	0.68	5.80	1.32
p-value				0.2327		0.9627		0.7849	

There was no significant difference in the number of *Lactobacillus* and *Bacillus* at the sampling time in both two groups. Regarding to the amount of *E. coli* before supplement, the number of *E. coli* in the treatment group was higher than that in the control group. However, after 4 weeks of treatment, the number of *E. coli* found in the experimental group was significantly decreasing to be equal to the control group. Bacterial numbers of *Lactobacillus*, *E. coli*, *Bacillus* in pullet, layer, before and after can be seen in Figure 9-11. Thus, using MICRO – COMPLETE can improve microflora in chicken gut.

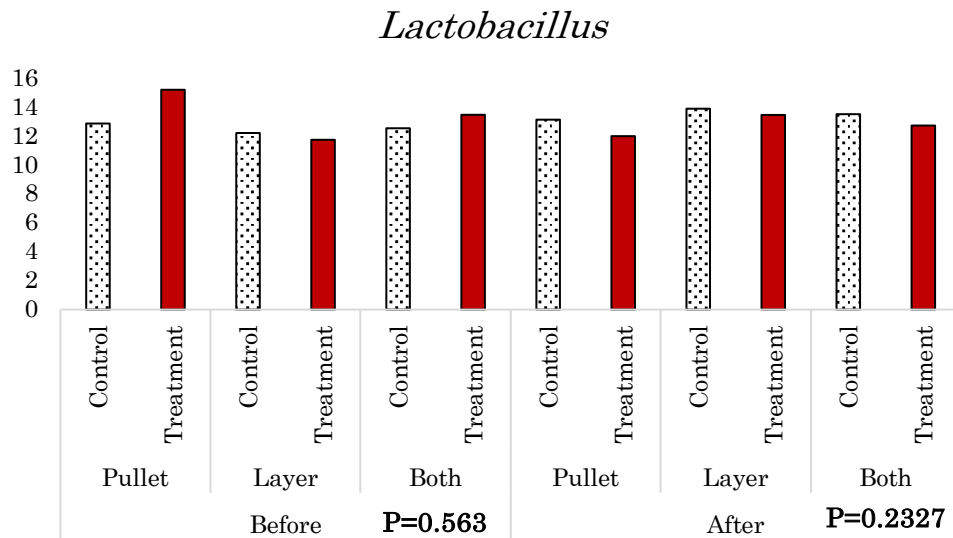


Figure 9. Number of *Lactobacillus* (logarite scale) in a gram feces before/after supplement in 2 types of chickens from 2 groups

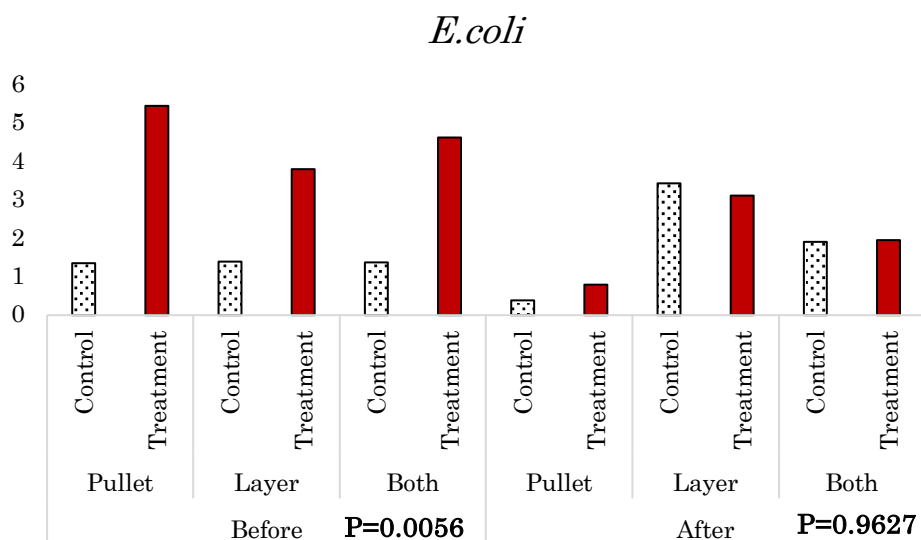


Figure 10. Number of *E. coli* (logarite scale) in a gram feces before/after supplement in 2 types of chickens from 2 groups

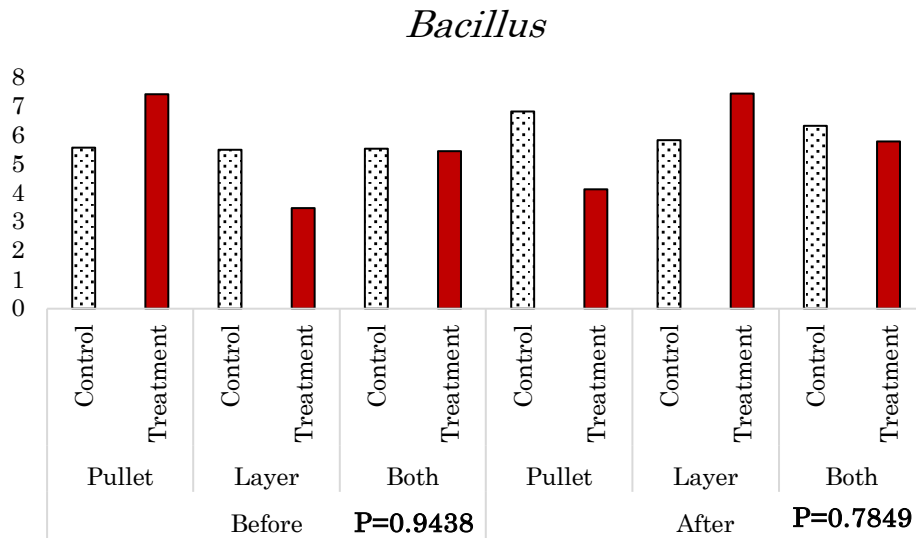


Figure 11. Number of *Bacillus* (logarithmic scale) in a gram feces before/after supplement in 2 types of chickens from 2 groups

- *Villus height and crypt depth in the small intestine*

After PROBYN supplementing 4 weeks, the length of villi in small intestine (duodenum section) was improved. It was seen before supplement, the length in treatment group was shorter significantly but after that it was improving to be the same with control group. This pattern was also seen in the crypt depth. Table 9, Figures 12 and 13 show this pattern. It proved that using MICRO – COMPLETE can improve villus height and crypt depth in the small intestine of chicken.

Table 9. Villus height and crypt depth in the small intestine of chicken before/after supplement in 2 types of chicken and 2 groups

			N	Villus height		Crypt depth	
				micrometer	SD	micrometer	SD
Before	Pullet	Control	5	1321.6	138.8	264.6	30.5
		Treatment	5	1118.4	53.9	201.2	27.5
	Layer	Control	5	1347	53.7	299.0	38.3
		Treatment	5	1259	319.4	201.8	61.1
	Both	Control	10	1334.3	100.1	281.8	37.3
		Treatment	10	1188.7	228.3	201.5	44.7
After	Pullet	Control	5	1175.4	278.5	239.0	62.4
		Treatment	5	1323	93.5	265.8	146.6
	Layer	Control	5	1257.8	110.9	272.0	80.2
		Treatment	5	1240.2	160.5	207.4	36.9
	Both	Control	10	1216.6	204.5	255.5	69.9
		Treatment	10	1281.6	131.3	236.6	105.4

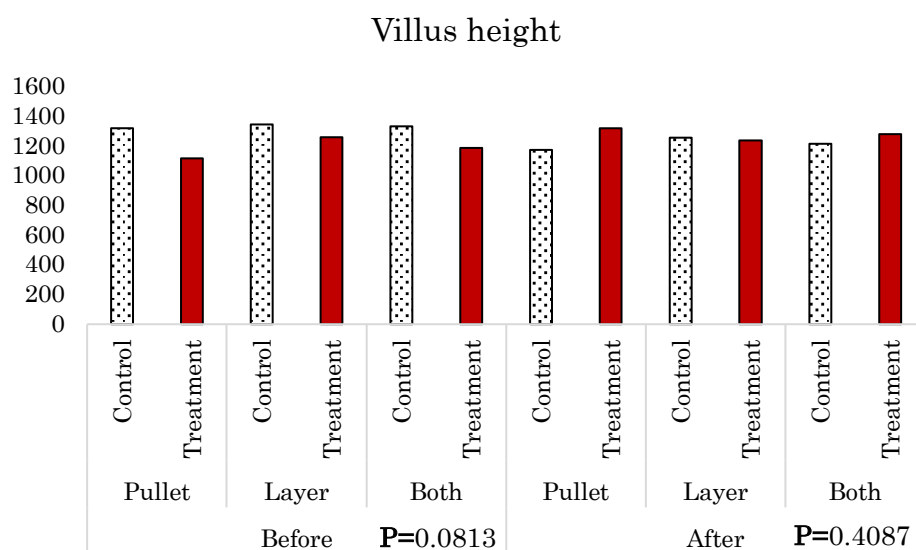


Figure 12. Villus height in the small intestine of chicken before/after supplement in 2 types of chicken and 2 groups

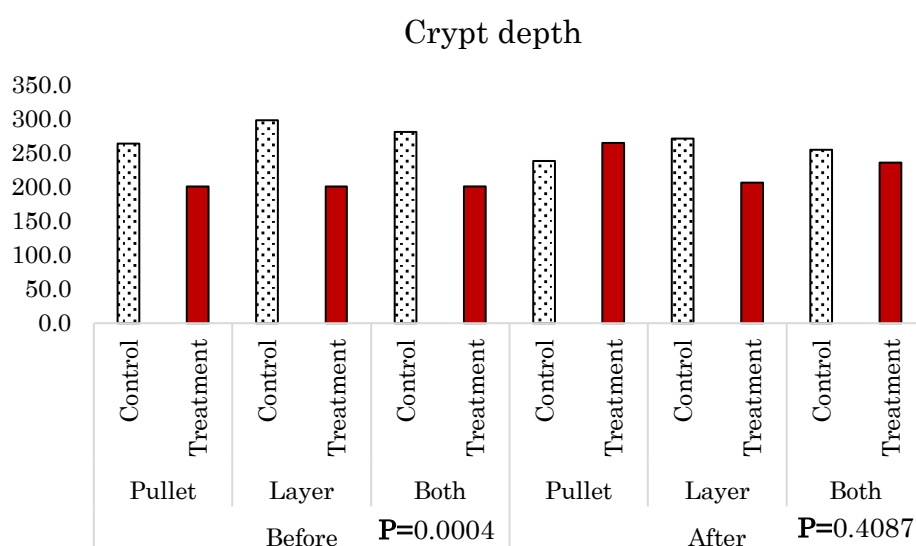


Figure 12. Crypt depth in the small intestine of chicken before/after supplement in 2 types of chicken and 2 groups

Conclusions

MICRO – COMPLETE supplement for pullets and layers could show some benefits on by

- *Higher egg production*
- *Better fecal score*
- *Improve intestinal microflora data by reduction of *E.coli**
- *Improve villi height and Crypt depth of gut.*

Thus, this product is recommended to use on chicken farms.

Appendix – Statistical output

```
. * Fecal score
. use "D:\workplace\project\NL group\PROBYN - BRENNTAG\moi\diemphan.dta", clear
. tab type
```

type	Freq.	Percent	Cum.
layer	2,240	33.33	33.33
pullet	4,480	66.67	100.00

```
. tab type breed
```

type	breed			Total
	HYLINE	ISA BOV..	LOHMANN..	
layer	0	1,120	1,120	2,240
pullet	2,240	0	2,240	4,480
Total	2,240	1,120	3,360	6,720

```
. tab fecal_x treatment
```

fecal_x	treatment		Total
	1	2	
0	49	195	244
1	2,250	2,437	4,687
2	898	662	1,560
3	163	66	229
Total	3,360	3,360	6,720

```
. tab fecal_x treatment if type=="pullet"
```

fecal_x	treatment		Total
	1	2	
0	0	1	1
1	1,729	1,824	3,553
2	465	379	844
3	46	36	82
Total	2,240	2,240	4,480

```
. tab fecal_x treatment if type=="ga_de"
```

fecal_x	treatment		Total
	1	2	
0	49	194	243
1	521	613	1,134
2	433	283	716
3	117	30	147
Total	1,120	1,120	2,240

```
. * So sanh diem phan
. kwallis fecal_x if type=="pullet", by(treatment)
```

Kruskal-Wallis equality-of-populations rank test

treatm~t	Obs	Rank Sum
1	2,240	5.13e+06
2	2,240	4.91e+06

```
chi-squared = 6.310 with 1 d.f.
probability = 0.0120
```

```
chi-squared with ties = 12.761 with 1 d.f.
probability = 0.0004
```

```
. kwallis fecal_x if type=="ga_de", by(treatment)
```

```
Kruskal-Wallis equality-of-populations rank test
```

```
+-----+
| treatm~t |  Obs | Rank Sum |
+-----+
|           |  1 | 1,120 | 1.43e+06 |
|           |  2 | 1,120 | 1.08e+06 |
+-----+
```

```
chi-squared = 135.706 with 1 d.f.
probability = 0.0001
```

```
chi-squared with ties = 162.321 with 1 d.f.
probability = 0.0001
```

```
. * Fecal score by time
. sort age
. by age: sum fecal_x if type=="pullet" & treatment==1
```

```
-----
-> age = 15
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      480   1.229167   .5063102     1       3
-----
-> age = 16
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      560   1.335714   .5229709     1       3
-----
-> age = 17
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      560   1.214286   .4277614     1       3
-----
-> age = 18
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      560   1.196429   .4237849     1       3
-----
-> age = 19
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |       80   1.3625    .556748     1       3
-----
```

```
. sort age
. by age: sum fecal_x if type=="pullet" & treatment==2
```

```
-----
-> age = 15
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      480   1.189583   .4608876     0       3
-----
-> age = 16
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      560   1.285714   .5080492     1       3
-----
-> age = 17
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      560   1.1625    .3927166     1       3
-----
-> age = 18
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |      560   1.160714   .377203     1       3
-----
-> age = 19
Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
fecal_x |       80   1.225    .4493314     1       3
-----
```

```
. sort age
```

```

. by age: sum fecal_x if type=="ga_de" & treatment==1
-> age = 24
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  fecal_x |      80     1.475   .6555508     0        3
-----+-----
-> age = 25
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  fecal_x |     280     1.617857  .6723167     0        3
-----+-----
-> age = 26
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  fecal_x |     280     1.578571  .7993533     0        3
-----+-----
-> age = 27
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  fecal_x |     280     1.389286  .6573757     0        3
-----+-----
-> age = 28
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  fecal_x |     200     1.68     .8312973     0        3
-----+-----
. * *****
. * Pullet weight
. * *****
. use "D:\workplace\project\NL group\PROBYN - BRENNTAG\moi\weight_pullet.dta", clear

. sort age
. by age: sum weight if treatment ==1
-----+-----
-> age = 15
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80     943.25   128.2381    660     1200
-----+-----
-> age = 16
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80    1085.25   105.8776    840     1300
-----+-----
-> age = 17
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80    1160.25    86.58025    930     1390
-----+-----
-> age = 18
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80    1179.5    109.8203    960     1470
-----+-----
-> age = 19
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80    1254.875   130.0145    910     1510
-----+-----
.
. sort age
. by age: sum weight if treatment ==2
-----+-----
-> age = 15
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80     980.75   109.2179    600     1200
-----+-----
-> age = 16
  Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
  weight |      80    1067.375   115.985     790     1290
-----+-----

```

```
-----
-> age = 17
  Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
  weight |      80   1128.375   105.3679     880    1360
-----
-> age = 18
  Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
  weight |      80   1184.988   107.9657     900    1400
-----
-> age = 19
  Variable |      Obs      Mean   Std. Dev.   Min     Max
-----+-----
  weight |      80   1247.75    108.9649     950    1480
-----
```

```
. oneway weight treatment if age==15, tabulate
```

treatment	Summary of weight		
	Mean	Std. Dev.	Freq.
1	943.25	128.23806	80
2	980.75	109.21788	80
Total	962	120.21365	160

Source	Analysis of Variance			
	SS	df	MS	Prob > F
Between groups	56250	1	56250	3.96
Within groups	2241510	158	14186.7722	0.0482
Total	2297760	159	14451.3208	

```
Bartlett's test for equal variances:  chi2(1) = 2.0147  Prob>chi2 = 0.156
```

```
. oneway weight treatment if age==16, tabulate
```

treatment	Summary of weight		
	Mean	Std. Dev.	Freq.
1	1085.25	105.87759	80
2	1067.375	115.98498	80
Total	1076.3125	111.05906	160

Source	Analysis of Variance			
	SS	df	MS	Prob > F
Between groups	12780.625	1	12780.625	1.04
Within groups	1948343.75	158	12331.2896	0.3102
Total	1961124.38	159	12334.1156	

```
Bartlett's test for equal variances:  chi2(1) = 0.6517  Prob>chi2 = 0.420
```

```
. oneway weight treatment if age==17, tabulate
```

treatment	Summary of weight		
	Mean	Std. Dev.	Freq.
1	1160.25	86.580247	80
2	1128.375	105.36788	80
Total	1144.3125	97.44937	160

Source	Analysis of Variance			
	SS	df	MS	Prob > F
Between groups	40640.625	1	40640.625	4.37
Within groups	1469283.75	158	9299.26424	0.0382
Total	1509924.38	159	9496.37972	

```
Bartlett's test for equal variances:  chi2(1) = 3.0084  Prob>chi2 = 0.083
```

```
. oneway weight treatment if age==18, tabulate
```


treatment	Summary of weight		
	Mean	Std. Dev.	Freq.
1	1179.5	109.82034	80
2	1184.9875	107.96571	80
Total	1182.2438	108.58888	160

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	1204.50625	1	1204.50625	0.10	0.7504
Within groups	1873650.99	158	11858.5506		
Total	1874855.49	159	11791.544		

Bartlett's test for equal variances: $\chi^2(1) = 0.0228$ Prob> $\chi^2 = 0.880$

. oneway weight treatment if age==19, tabulate

treatment	Summary of weight		
	Mean	Std. Dev.	Freq.
1	1254.875	130.01454	80
2	1247.75	108.96492	80
Total	1251.3125	119.62794	160

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	2030.625	1	2030.625	0.14	0.7077
Within groups	2273393.75	158	14388.568		
Total	2275424.38	159	14310.8451		

Bartlett's test for equal variances: $\chi^2(1) = 2.4363$ Prob> $\chi^2 = 0.119$

.
.
. oneway weight treatment , tabulate

treatment	Summary of weight		
	Mean	Std. Dev.	Freq.
1	1124.625	154.4701	400
2	1121.8475	143.02125	400
Total	1123.2362	148.76909	800

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	1542.90125	1	1542.90125	0.07	0.7919
Within groups	17682119.4	798	22158.0444		
Total	17683662.3	799	22132.2432		

Bartlett's test for equal variances: $\chi^2(1) = 2.3608$ Prob> $\chi^2 = 0.124$

. *****
. * Production parameter
. *****
.
. use "D:\workplace\project\NL group\PROBYN - BRENNTAG\moi\nangsuat.dta", clear

treatment	type		Total
	pullet	layer	
1	56	42	98
2	56	42	98
Total	112	84	196

. gen mortality = dead*1000/ number

```
. sum mortality
```

Variable	Obs	Mean	Std. Dev.	Min	Max
mortality	196	.3067538	.6988599	0	9.588493

```
. * Pullet
```

```
. oneway feed_chick treatment if type=="pullet"
```

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	.012721986	1	.012721986	0.00	0.9880
Within groups	6186.89478	110	56.244498		
Total	6186.9075	111	55.7379054		

```
Bartlett's test for equal variances: chi2(1) = 6.8149 Prob>chi2 = 0.009
```

```
. sort age
```

```
. by age: sum feed_chick if type=="pullet" & treatment==1
```

```
-----
```

-> age = 15					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	12	61.95608	6.088112	51.36716	67.9565

```
-----
```

-> age = 16					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	14	69.85103	4.680209	61.83182	82.13799

```
-----
```

-> age = 17					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	14	65.38815	4.987836	58.05573	72.16164

```
-----
```

-> age = 18					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	14	55.0302	10.5595	38.0548	72.3589

```
-----
```

-> age = 19					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	2	60.23115	5.825034	56.11222	64.35007

```
-----
```

```
. sort age
```

```
. by age: sum feed_chick if type=="pullet" & treatment==2
```

```
-----
```

-> age = 15					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	12	58.40807	2.785887	54.54262	62.4171

```
-----
```

-> age = 16					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	14	66.40149	3.19261	60.75572	72.27127

```
-----
```

-> age = 17					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	14	62.0185	5.791643	54.74738	72.28822

```
-----
```

-> age = 18					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	14	63.75837	7.625924	55.0228	76.46459

```
-----
```

-> age = 19					
Variable	Obs	Mean	Std. Dev.	Min	Max
feed_chick	2	68.7531	10.96509	60.99961	76.50659

```
-----
```

```
. ** feed/bird/day
. xi: mixed feed_chick i.treatment || day: if type=="pullet" & age==15
i.treatment      _Itreatment_1-2      (naturally coded; _Itreatment_1 omitted)
Performing EM optimization:
Performing gradient-based optimization:
```

```
Iteration 0:  log likelihood = -67.337566
Iteration 1:  log likelihood = -67.337566
```

Computing standard errors:

```
Mixed-effects ML regression      Number of obs      =      24
Group variable: day              Number of groups    =       6

Obs per group:
    min =      4
    avg  =     4.0
    max  =      4

Wald chi2(1) =      6.85
Prob > chi2   =     0.0089

Log likelihood = -67.337566
```

feed_chick	Coef.	Std. Err.	z	P> z	[95% Conf. Interval]	
_Itreatment_2	-3.548011	1.355503	-2.62	0.009	-6.204748	-.891274
_cons	61.95608	1.582889	39.14	0.000	58.85368	65.05849

Random-effects Parameters	Estimate	Std. Err.	[95% Conf. Interval]	
day: Identity				
var(_cons)	9.521052	7.147496	2.186171	41.46539
var(Residual)	11.02433	3.674777	5.736122	21.18781

LR test vs. linear model: chibar2(01) = 5.98 Prob >= chibar2 = 0.0072

```
. xi: mixed feed_chick i.treatment || day: if type=="pullet" & age==16
i.treatment      _Itreatment_1-2      (naturally coded; _Itreatment_1 omitted)
```

Performing EM optimization:

Performing gradient-based optimization:

```
Iteration 0:  log likelihood = -77.107231
Iteration 1:  log likelihood = -77.106982
Iteration 2:  log likelihood = -77.106982
```

Computing standard errors:

```
Mixed-effects ML regression      Number of obs      =      28
Group variable: day              Number of groups    =       7

Obs per group:
    min =      4
    avg  =     4.0
    max  =      4

Wald chi2(1) =      6.63
Prob > chi2   =     0.0100

Log likelihood = -77.106982
```

feed_chick	Coef.	Std. Err.	z	P> z	[95% Conf. Interval]	
_Itreatment_2	-3.449532	1.339353	-2.58	0.010	-6.074615	-.8244482
_cons	69.85103	1.109936	62.93	0.000	67.67559	72.02646

Random-effects Parameters	Estimate	Std. Err.	[95% Conf. Interval]	
day: Identity				
var(_cons)	2.345167	3.087486	.1776399	30.96045
var(Residual)	12.55706	3.875193	6.858065	22.99188


```
-----+-----
day: Identity |
      var(_cons) | 6.17e-22  9.57e-21  3.85e-35  9.87e-09
-----+-----
      var(Residual) | 78.76966  21.05216  46.65142  133.0004
-----+-----
```

LR test vs. linear model: chibar2(01) = 0.00 Prob >= chibar2 = 1.0000

```
. xi: mixed feed_chick i.treatment || day: if type=="pullet" & age==19
i.treatment      _Itreatment_1-2      (naturally coded; _Itreatment_1 omitted)
```

Performing EM optimization:

Performing gradient-based optimization:

```
Iteration 0:  log likelihood = -13.003432
Iteration 1:  log likelihood = -12.979481
Iteration 2:  log likelihood = -12.979203
Iteration 3:  log likelihood = -12.979203
```

Computing standard errors:

```
Mixed-effects ML regression      Number of obs      =      4
Group variable: day              Number of groups   =      1

Obs per group:
      min =      4
      avg =     4.0
      max =      4

Wald chi2(1) =      1.88
Prob > chi2  =     0.1698

Log likelihood = -12.979203
```

```
-----+-----
feed_chick |      Coef.  Std. Err.      z    P>|z|    [95% Conf. Interval]
-----+-----
_Itreatment_2 | 8.521954  6.208146    1.37  0.170   -3.645789   20.6897
  _cons | 60.23115  4.389822   13.72  0.000   51.62725   68.83504
-----+-----
```

```
-----+-----
Random-effects Parameters |      Estimate  Std. Err.    [95% Conf. Interval]
-----+-----
day: Identity
      var(_cons) | 6.38e-22  3.31e-20   3.94e-66   1.03e+23
-----+-----
      var(Residual) | 38.54108  27.25575   9.637519  154.1283
-----+-----
```

LR test vs. linear model: chibar2(01) = 0.00 Prob >= chibar2 = 1.0000

```
.
. ** ti lệ chết
. sort age

. by age: sum mortality if type=="pullet" & treatment==1
```

-> age = 15

```
-----+-----
Variable |      Obs      Mean  Std. Dev.    Min      Max
-----+-----
mortality |      12  1.196435  2.648763  .1580528  9.588493
-----+-----
```

-> age = 16

```
-----+-----
Variable |      Obs      Mean  Std. Dev.    Min      Max
-----+-----
mortality |      14  .439777  .3486647     0  1.438159
-----+-----
```

-> age = 17

```
-----+-----
Variable |      Obs      Mean  Std. Dev.    Min      Max
-----+-----
mortality |      14  .2004355  .2008843     0  .7224274
-----+-----
```

-> age = 18

```

Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |       14   .1662466   .1589028     0   .4823927
-----+-----
-> age = 19
Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |        2   .2413127   .3412678     0   .4826255
-----+-----
. sort age

. by age: sum mortality if type=="pullet" & treatment==2
-----+-----
-> age = 15
Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |       12   .5145017   .3426951   .1560062   1.4068
-----+-----
-> age = 16
Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |       14   .2573365   .2072335     0   .784683
-----+-----
-> age = 17
Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |       14   .2069981   .1906118     0   .6261251
-----+-----
-> age = 18
Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |       14   .207471   .2359667     0   .7869678
-> age = 19
Variable |      Obs      Mean   Std. Dev.   Min      Max
-----+-----
mortality |        2   .3533482   .2770867   .1574183   .5492781
-----+-----
. xi: mixed mortality i.treatment || day: if type=="pullet" & age==15
i.treatment      _Itreatment_1-2      (naturally coded; _Itreatment_1 omitted)

Performing EM optimization:

Performing gradient-based optimization:

Iteration 0:  log likelihood = -48.369839
Iteration 1:  log likelihood = -48.270052
Iteration 2:  log likelihood = -48.27005

Computing standard errors:

Mixed-effects ML regression              Number of obs   =          24
Group variable: day                     Number of groups =           6

Obs per group:
      min =          4
      avg =          4.0
      max =          4

Wald chi2(1) =          0.85
Prob > chi2  =          0.3556

Log likelihood = -48.27005

-----+-----
mortality |      Coef.   Std. Err.   z   P>|z|   [95% Conf. Interval]
-----+-----
_Itreatment_2 |  -.6819335   .738181   -0.92  0.356   -2.128742   .7648748
  _cons |   1.196435   .5219728   2.29  0.022   .1733872   2.219483
-----+-----

Random-effects Parameters | Estimate   Std. Err.   [95% Conf. Interval]
-----+-----
day: Identity
      var(_cons) |   1.22e-15   2.24e-14   2.59e-31   5.710322

```

```
-----+-----
var(Residual) | 3.269468 .943814 1.856762 5.75702
-----+-----
LR test vs. linear model: chibar2(01) = 0.00 Prob >= chibar2 = 1.0000
```

```
. xi: mixed mortality i.treatment || day: if type=="pullet" & age==16
i.treatment _Itreatment_1-2 (naturally coded; _Itreatment_1 omitted)
```

Performing EM optimization:

Performing gradient-based optimization:

```
Iteration 0: log likelihood = -3.784376
Iteration 1: log likelihood = -3.7220815
Iteration 2: log likelihood = -3.7219685
Iteration 3: log likelihood = -3.7219675
Iteration 4: log likelihood = -3.7219675
```

Computing standard errors:

```
Mixed-effects ML regression      Number of obs   =      28
Group variable: day              Number of groups =       7

Obs per group:
      min =      4
      avg =     4.0
      max =      4

Wald chi2(1) =      3.05
Prob > chi2   =     0.0807

Log likelihood = -3.7219675
```

```
-----+-----
mortality |      Coef.   Std. Err.      z    P>|z|     [95% Conf. Interval]
-----+-----
_Itreatment_2 | -1.1824405   .1044584   -1.75   0.081   -1.3871753   -.0222942
  _cons | .439777     .0738633    5.95   0.000    .2950077    .5845463
-----+-----
```

```
-----+-----
Random-effects Parameters | Estimate Std. Err.   [95% Conf. Interval]
-----+-----
day: Identity
      var(_cons) | 2.66e-17  4.06e-16   2.65e-30   .000267
-----+-----
      var(Residual) | .0763809  .0204138   .0452367   .1289672
-----+-----
```

```
LR test vs. linear model: chibar2(01) = 0.00 Prob >= chibar2 = 1.0000
```

```
. xi: mixed mortality i.treatment || day: if type=="pullet" & age==17
i.treatment _Itreatment_1-2 (naturally coded; _Itreatment_1 omitted)
```

Performing EM optimization:

Performing gradient-based optimization:

```
Iteration 0: log likelihood = 7.3301877
Iteration 1: log likelihood = 7.3306834
Iteration 2: log likelihood = 7.3306834
```

Computing standard errors:

```
Mixed-effects ML regression      Number of obs   =      28
Group variable: day              Number of groups =       7

Obs per group:
      min =      4
      avg =     4.0
      max =      4

Wald chi2(1) =      0.01
Prob > chi2   =     0.9209

Log likelihood = 7.3306834
```

```
-----+-----
mortality |      Coef.   Std. Err.      z    P>|z|     [95% Conf. Interval]
-----+-----
_Itreatment_2 | .0065626   .0660529    0.10   0.921   -1.1228987   .1360239
  _cons | .2004355   .0538973    3.72   0.000    .0947988    .3060722
-----+-----
```

```

-----
-----
Random-effects Parameters | Estimate Std. Err. [95% Conf. Interval]
-----+-----
day: Identity            |
      var(_cons)         | .005064 .0071853 .0003138 .0817097
-----+-----
      var(Residual)      | .0305409 .0094251 .01668 .0559201
-----+-----
LR test vs. linear model: chibar2(01) = 0.73          Prob >= chibar2 = 0.1958

. xi: mixed mortality i.treatment || day: if type=="pullet" & age==18
i.treatment      _Itreatment_1-2 (naturally coded; _Itreatment_1 omitted)

Performing EM optimization:

Performing gradient-based optimization:

Iteration 0:  log likelihood = 9.478674
Iteration 1:  log likelihood = 9.478674

Computing standard errors:

Mixed-effects ML regression              Number of obs   =      28
Group variable: day                     Number of groups =       7

Obs per group:
      min =      4
      avg =     4.0
      max =      4

Wald chi2(1) =      0.57
Prob > chi2   =     0.4485

-----
-----
mortality |      Coef.  Std. Err.      z    P>|z|    [95% Conf. Interval]
-----+-----
_Itreatment_2 | .0412244 .0543953   0.76  0.449   -.0653884 .1478371
  _cons | .1662466 .0623571   2.67  0.008   .0440289 .2884643
-----+-----

-----
-----
Random-effects Parameters | Estimate Std. Err. [95% Conf. Interval]
-----+-----
day: Identity            |
      var(_cons)         | .0168629 .0118892 .0042344 .0671543
-----+-----
      var(Residual)      | .0207119 .0063918 .0113118 .0379233
-----+-----
LR test vs. linear model: chibar2(01) = 6.54          Prob >= chibar2 = 0.0053

. xi: mixed mortality i.treatment || day: if type=="pullet" & age==19
i.treatment      _Itreatment_1-2 (naturally coded; _Itreatment_1 omitted)

Performing EM optimization:

Performing gradient-based optimization:

Iteration 0:  log likelihood = .36024314
Iteration 1:  log likelihood = .3767805
Iteration 2:  log likelihood = .38446877
Iteration 3:  log likelihood = .38447195
Iteration 4:  log likelihood = .38447195

Computing standard errors:

Mixed-effects ML regression              Number of obs   =       4
Group variable: day                     Number of groups =      1

Obs per group:
      min =       4
      avg =     4.0
      max =       4

Wald chi2(1) =      0.26
Prob > chi2   =     0.6102
-----

```



```

    feed_chick |           7    115.6173    1.213996    114.6023    116.9317
-----+-----
-> age = 27
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    112.7311    2.577562    110.3482    116.9838
-----+-----
-> age = 28
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    112.7218    .0587439    112.6478    112.8088
-----+-----
-> age = 29
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    114.8359    2.445151    112.824    117.4759
-----+-----
-> age = 30
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           5    117.0881    .5913796    116.4405    117.5443

. sum feed_chick if type=="layer" & treatment==1

  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           42    113.9619    2.431764    110.0587    117.5443
-> age = 24
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           2    115.4299    .0181211    115.4171    115.4427
-----+-----
-> age = 25
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    115.5563    .0562106    115.4683    115.6326
-----+-----
-> age = 26
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    115.7393    .0574243    115.6584    115.8129
-----+-----
-> age = 27
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    115.8992    .0484394    115.8336    115.9627
-----+-----
-> age = 28
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    116.0626    .0641438    115.9886    116.1596
-----+-----
-> age = 29
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           7    116.2553    .0639399    116.1752    116.3467
-----+-----
-> age = 30
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           5    116.4364    .0556084    116.3727    116.5136

. sum feed_chick if type=="layer" & treatment==2
  Variable |           Obs           Mean    Std. Dev.           Min           Max
-----+-----
    feed_chick |           42    115.9436    .3120291    115.4171    116.5136

.
. xi: mixed feed_chick i.treatment || day: if type=="layer"
i.treatment      _Itreatment_1-2 (naturally coded; _Itreatment_1 omitted)

Performing EM optimization:

Performing gradient-based optimization:

Iteration 0:  log likelihood = -164.24238
Iteration 1:  log likelihood = -164.13562

```



```

. sort age

. by age: sum mortality if type=="layer" & treatment==1
-> age = 24
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         2   .3999734   .0001131   .3998934   .4000534
-----+-----
-> age = 25
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .1207386   .1017847         0   .2668683
-----+-----
-> age = 26
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .1781168   .0574677   .0890115   .2671178
-----+-----
-> age = 27
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .1655943   .055867   .0891226   .2673916
-----+-----
-> age = 28
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .2232115   .0815082   .1338688   .357095
-----+-----
-> age = 29
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .2491318   .050569   .179011   .3128771
-----+-----
-> age = 30
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         5   .1253586   .0861069         0   .2238438

. sum mortality if type=="layer" & treatment==1
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |        42   .1901021   .0944264         0   .4000534

. sort age

. by age: sum mortality if type=="layer" & treatment==2
-> age = 24
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         2   .2219805   .0000348   .2219559   .2220051
-----+-----
-> age = 25
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .2349063   .0756423   .1777146   .399698
-----+-----
-> age = 26
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .2162043   .047524   .1781103   .3115265
-----+-----
-> age = 27
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .1910417   .0714952   .0891941   .312082
-----+-----
-> age = 28
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .2295828   .0871525   .089222   .3126396
-----+-----
-> age = 29
  Variable |      Obs      Mean   Std. Dev.      Min      Max
-----+-----
 mortality |         7   .2427391   .0507204   .1340483   .2684203

```

```

-----
-> age = 30
  Variable |          Obs          Mean    Std. Dev.    Min          Max
-----+-----
  mortality |             5    .2955806    .0679807    .2237937    .4031536

. sum mortality if type=="layer" & treatment==2

  Variable |          Obs          Mean    Std. Dev.    Min          Max
-----+-----
  mortality |            42    .2315044    .0685824    .0891941    .4031536

. xi: mepoisson dead i.treatment i.age if type=="layer", exposure(number) || house:

Mixed-effects Poisson regression
Group variable:      house

                                Number of obs   =      84
                                Number of groups =       2

                                Obs per group:
                                    min =      42
                                    avg  =     42.0
                                    max  =      42

Integration method: mvaghermite                                Integration pts. =       7

                                Wald chi2(7)      =     13.63
Log likelihood = -168.81188                                Prob > chi2   =     0.0581
-----
          dead |          Coef.    Std. Err.    z    P>|z|    [95% Conf. Interval]
-----+-----
  _Itreatment_2 |    .1968145    .100865    1.95  0.051    -.0008773    .3945063
  _Iage_16 |           0 (omitted)
  _Iage_17 |           0 (omitted)
  _Iage_18 |           0 (omitted)
  _Iage_19 |           0 (omitted)
  _Iage_24 |    .3902035    .2387276    1.63  0.102    -.0776941    .858101
  _Iage_25 |   -.1683544    .1978225   -0.85  0.395    -.5560794    .2193706
  _Iage_26 |   -.06527    .1934053   -0.34  0.736    -.4443375    .3137976
  _Iage_27 |   -.1657491    .1978225   -0.84  0.402    -.553474    .2219759
  _Iage_28 |    .0729502    .1880455    0.39  0.698    -.2956122    .4415125
  _Iage_29 |    .1557726    .1851043    0.84  0.400    -.2070252    .5185704
  _Iage_30 |           0 (omitted)
  _cons |   -8.569433    .1560225  -54.92  0.000    -8.875231   -8.263635
  ln(number) |           1 (exposure)
-----+-----
house
  var(_cons) |    9.44e-34    3.18e-18
-----+-----
LR test vs. Poisson model: chi2(0) = 1.7e-13                                Prob > chi2 = .

Note: LR test is conservative and provided only for reference.

.
. * Nang suất trung
. gen egg_18_n= egg_18 * 300
(112 missing values generated)
. gen egg_19_n= egg_19 * 300
(112 missing values generated)
. gen egg_20_n= egg_20 * 300
(112 missing values generated)
. gen egg_other_n= egg_other * 30
(112 missing values generated)
. gen egg_n = egg_18_n + egg_19_n + egg_20_n + egg_other_n
(112 missing values generated)
. *br egg_n egg
. gen egg_chick = egg_n/ number
(112 missing values generated)
. sum egg_chick

  Variable |          Obs          Mean    Std. Dev.    Min          Max
-----+-----
  egg_chick |            84    .9259671    .0258288    .839776    1.028862

. xi: mixed egg_chick i.treatment || day: if type=="layer"
i.treatment      _Itreatment_1-2    (naturally coded; _Itreatment_1 omitted)
Performing EM optimization:

Performing gradient-based optimization:

```



```

. sort age

. by age:sum egg_chick if treatment==2

-> age = 24
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         2   .8617288   .005786   .8576375   .8658201
-----+-----
-> age = 25
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         7   .8969626   .0122265   .8793356   .9072715
-----+-----
-> age = 26
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         7   .9218444   .0011292   .9208185   .9241865
-----+-----
-> age = 27
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         7   .9359199   .0058035   .9275595   .943312
-----+-----
-> age = 28
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         7   .9441244   .0005218   .9435225   .9449136
-----+-----
-> age = 29
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         7   .9456924   .0005201   .9450402   .9464357
-----+-----
-> age = 30
Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |         5   .9452828   .0038681   .9383823   .9474109

```

```

. sum egg_chick if treatment==2

Variable |      Obs      Mean   Std. Dev.   Min       Max
-----+-----
egg_chick |        42   .927659   .023651   .8576375   .9474109

```

```

.
. xi: poisson egg_n i.treatment i.age, exposure(number)
i.treatment      _Itreatment_1-2 (naturally coded; _Itreatment_1 omitted)
i.age            _Iage_15-30 (naturally coded; _Iage_15 omitted)
note: _Iage_16 omitted because of collinearity
note: _Iage_17 omitted because of collinearity
note: _Iage_18 omitted because of collinearity
note: _Iage_19 omitted because of collinearity
note: _Iage_30 omitted because of collinearity

```

```

Iteration 0:  log likelihood = -691.85076
Iteration 1:  log likelihood = -691.85076

```

```

Poisson regression                               Number of obs   =          84
                                                LR chi2(7)      =       958.41
                                                Prob > chi2     =       0.0000
Log likelihood = -691.85076                     Pseudo R2      =       0.4092

```

egg_n	Coef.	Std. Err.	z	P> z	[95% Conf. Interval]	
_Itreatment_2	.0036315	.0015145	2.40	0.016	.0006631	.0065998
_Iage_16	0	(omitted)				
_Iage_17	0	(omitted)				
_Iage_18	0	(omitted)				
_Iage_19	0	(omitted)				
_Iage_24	-.0915558	.0042171	-21.71	0.000	-.0998212	-.0832903
_Iage_25	-.0411493	.0028843	-14.27	0.000	-.0468025	-.0354961
_Iage_26	-.0044497	.0028629	-1.55	0.120	-.0100609	.0011615
_Iage_27	.0000142	.002861	0.00	0.996	-.0055933	.0056217
_Iage_28	.0033183	.0028599	1.16	0.246	-.0022869	.0089236

```

    _Iage_29 | .0016524 .0028619 0.58 0.564 -.0039568 .0072616
    _Iage_30 | 0 (omitted)
    _cons | -.0678839 .002315 -29.32 0.000 -.0724212 -.0633466
ln(number) | 1 (exposure)

```

```

-----
. *****
. * LAB
. *****
. use "D:\workplace\project\NL group\PROBYN - BRENNTAG\moi\lab.dta", clear
. sum lab clos ecoli bacillus length depth

```

Variable	Obs	Mean	Std. Dev.	Min	Max
lab	40	4710925	9970650	1000	5.00e+07
clos	40	0	0	0	0
ecoli	40	211.684	566.2926	.03	2400
bacillus	40	5740.75	14208.67	0	80000
length	40	1255.3	176.8356	717	1566
depth	40	243.85	73.14354	117	515

```

. tab treatment

```

treatment	Freq.	Percent	Cum.
1	20	50.00	50.00
2	20	50.00	100.00
Total	40	100.00	

```

. tab time

```

time	Freq.	Percent	Cum.
0	20	50.00	50.00
1	20	50.00	100.00
Total	40	100.00	

```

. gen lab_lg=log(lab+1)

```

```

. gen ecoli_lg=log(ecoli+1)

```

```

. gen baci_lg=log(bacillus+1)

```

```

. oneway lab_lg treatment if time==0 & type== "pullet", tabulate

```

treatment	Summary of lab_lg		
	Mean	Std. Dev.	Freq.
1	12.894509	3.8136809	5
2	15.233546	3.390111	5
Total	14.064027	3.6182531	10

Source	Analysis of Variance				
	SS	df	MS	F	Prob > F
Between groups	13.677739	1	13.677739	1.05	0.3354
Within groups	104.148058	8	13.0185072		
Total	117.825797	9	13.0917552		

Bartlett's test for equal variances: $\chi^2(1) = 0.0492$ Prob> $\chi^2 = 0.825$

```

. oneway lab_lg treatment if time==0 & type== "ga_de", tabulate

```

treatment	Summary of lab_lg		
	Mean	Std. Dev.	Freq.
1	12.237676	3.3591332	5
2	11.767311	3.4178459	5
Total	12.002493	3.2044221	10

Source	Analysis of Variance				
	SS	df	MS	F	Prob > F
Between groups	.553106176	1	.553106176	0.05	0.8318

Within groups	91.8617847	8	11.4827231
Total	92.4148909	9	10.2683212

Bartlett's test for equal variances: $\chi^2(1) = 0.0011$ Prob> $\chi^2 = 0.974$

. oneway lab_lg treatment if time==0 , tabulate

treatment	Summary of lab_lg		
	Mean	Std. Dev.	Freq.
1	12.566092	3.4057194	10
2	13.500429	3.6928617	10
Total	13.03326	3.4905157	20

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	4.3649241	1	4.3649241	0.35	0.5637
Within groups	227.12537	18	12.6180761		
Total	231.490294	19	12.1836997		

Bartlett's test for equal variances: $\chi^2(1) = 0.0558$ Prob> $\chi^2 = 0.813$

. oneway lab_lg treatment if time==1 & type== "pullet", tabulate

treatment	Summary of lab_lg		
	Mean	Std. Dev.	Freq.
1	13.161964	1.5748127	5
2	12.019677	1.3903847	5
Total	12.590821	1.5244258	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	3.26204825	1	3.26204825	1.48	0.2587
Within groups	17.6528189	8	2.20660236		
Total	20.9148672	9	2.32387413		

Bartlett's test for equal variances: $\chi^2(1) = 0.0550$ Prob> $\chi^2 = 0.815$

. oneway lab_lg treatment if time==1 & type== "ga_de", tabulate

treatment	Summary of lab_lg		
	Mean	Std. Dev.	Freq.
1	13.921755	1.0991682	5
2	13.489073	1.3474382	5
Total	13.705414	1.1814811	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	.468036014	1	.468036014	0.31	0.5932
Within groups	12.0950425	8	1.51188031		
Total	12.5630785	9	1.39589761		

Bartlett's test for equal variances: $\chi^2(1) = 0.1465$ Prob> $\chi^2 = 0.702$

. oneway lab_lg treatment if time==1 , tabulate

treatment	Summary of lab_lg		
	Mean	Std. Dev.	Freq.
1	13.54186	1.3414764	10
2	12.754375	1.5052812	10
Total	13.148117	1.4453107	20

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	3.10066184	1	3.10066184	1.53	0.2327
Within groups	36.588874	18	2.03271522		
Total	39.6895359	19	2.08892294		

Bartlett's test for equal variances: $\chi^2(1) = 0.1129$ Prob> $\chi^2 = 0.737$

.
 . oneway ecoli_lg treatment if time==0 & type== "pullet", tabulate

Summary of ecoli_lg			
treatment	Mean	Std. Dev.	Freq.
1	1.3516375	1.2243815	5
2	5.442955	2.5278947	5
Total	3.3972962	2.8558836	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	41.8471966	1	41.8471966	10.61	0.0116
Within groups	31.5574459	8	3.94468074		
Total	73.4046425	9	8.15607139		

Bartlett's test for equal variances: $\chi^2(1) = 1.7248$ Prob> $\chi^2 = 0.189$

. oneway ecoli_lg treatment if time==0 & type== "ga_de", tabulate

Summary of ecoli_lg			
treatment	Mean	Std. Dev.	Freq.
1	1.3928231	2.0238123	5
2	3.7969872	3.2117797	5
Total	2.5949051	2.8303002	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	14.4500127	1	14.4500127	2.01	0.1945
Within groups	57.6453808	8	7.2056726		
Total	72.0953934	9	8.01059927		

Bartlett's test for equal variances: $\chi^2(1) = 0.7329$ Prob> $\chi^2 = 0.392$

. oneway ecoli_lg treatment if time==0 , tabulate

Summary of ecoli_lg			
treatment	Mean	Std. Dev.	Freq.
1	1.3722303	1.5770558	10
2	4.6199711	2.8596065	10
Total	2.9961007	2.7977359	20

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	52.7391012	1	52.7391012	9.89	0.0056
Within groups	95.9800923	18	5.33222735		
Total	148.719193	19	7.82732597		

Bartlett's test for equal variances: $\chi^2(1) = 2.8567$ Prob> $\chi^2 = 0.091$

. oneway ecoli_lg treatment if time==1 & type== "pullet", tabulate

Summary of ecoli_lg			
treatment	Mean	Std. Dev.	Freq.

1	.38489054	.52424818	5
2	.79367654	1.7086192	5
Total	.58928354	1.2108137	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	.417764992	1	.417764992	0.26	0.6229
Within groups	12.7768629	8	1.59710787		
Total	13.1946279	9	1.46606977		

Bartlett's test for equal variances: $\chi^2(1) = 4.1123$ Prob> $\chi^2 = 0.043$

. oneway ecoli_lg treatment if time==1 & type== "ga_de", tabulate

Summary of ecoli_lg			
treatment	Mean	Std. Dev.	Freq.
1	3.4280105	1.9487536	5
2	3.109359	2.051187	5
Total	3.2686847	1.8936702	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	.253846972	1	.253846972	0.06	0.8075
Within groups	32.0200352	8	4.0025044		
Total	32.2738822	9	3.58598691		

Bartlett's test for equal variances: $\chi^2(1) = 0.0093$ Prob> $\chi^2 = 0.923$

. oneway ecoli_lg treatment if time==1, tabulate

Summary of ecoli_lg			
treatment	Mean	Std. Dev.	Freq.
1	1.9064505	2.0934117	10
2	1.9515177	2.1580072	10
Total	1.9289841	2.0693816	20

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	.010155282	1	.010155282	0.00	0.9627
Within groups	81.3543083	18	4.51968379		
Total	81.3644636	19	4.28234019		

Bartlett's test for equal variances: $\chi^2(1) = 0.0079$ Prob> $\chi^2 = 0.929$

.
.
.

. oneway baci_lg treatment if time==0 & type== "pullet", tabulate

Summary of baci_lg			
treatment	Mean	Std. Dev.	Freq.
1	5.5807405	3.2165649	5
2	7.4312957	1.0358898	5
Total	6.5060181	2.4549002	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	8.56138576	1	8.56138576	1.50	0.2556
Within groups	45.6774305	8	5.70967881		
Total	54.2388162	9	6.02653514		

Bartlett's test for equal variances: $\chi^2(1) = 3.8300$ Prob> $\chi^2 = 0.050$

. oneway baci_lg treatment if time==0 & type=="ga_de", tabulate

treatment	Summary of baci_lg		
	Mean	Std. Dev.	Freq.
1	5.5069458	.62577732	5
2	3.4857301	3.2342029	5
Total	4.4963379	2.4408547	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	10.2132825	1	10.2132825	1.88	0.2073
Within groups	43.4066625	8	5.42583282		
Total	53.619945	9	5.95777167		

Bartlett's test for equal variances: $\chi^2(1) = 7.0126$ Prob> $\chi^2 = 0.008$

. oneway baci_lg treatment if time==0 , tabulate

treatment	Summary of baci_lg		
	Mean	Std. Dev.	Freq.
1	5.5438432	2.1849272	10
2	5.4585129	3.0741081	10
Total	5.501178	2.5960784	20

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	.036406297	1	.036406297	0.01	0.9438
Within groups	128.016427	18	7.11202372		
Total	128.052833	19	6.73962281		

Bartlett's test for equal variances: $\chi^2(1) = 0.9752$ Prob> $\chi^2 = 0.323$

. oneway baci_lg treatment if time==1 & type=="pullet", tabulate

treatment	Summary of baci_lg		
	Mean	Std. Dev.	Freq.
1	6.8335362	3.9704928	5
2	4.1366161	3.7827656	5
Total	5.4850762	3.9225836	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	18.1834452	1	18.1834452	1.21	0.3035
Within groups	120.296514	8	15.0370643		
Total	138.47996	9	15.3866622		

Bartlett's test for equal variances: $\chi^2(1) = 0.0083$ Prob> $\chi^2 = 0.927$

. oneway baci_lg treatment if time==1 & type=="ga_de", tabulate

treatment	Summary of baci_lg		
	Mean	Std. Dev.	Freq.
1	5.8400364	5.4996778	5
2	7.4542084	4.2527576	5
Total	6.6471224	4.7122004	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	6.51387797	1	6.51387797	0.27	0.6177

Within groups	193.329616	8	24.166202
Total	199.843494	9	22.2048326

Bartlett's test for equal variances: $\chi^2(1) = 0.2325$ Prob> $\chi^2 = 0.630$

. oneway baci_lg treatment if time==1 , tabulate

treatment	Summary of baci_lg		
	Mean	Std. Dev.	Freq.
1	6.3367863	4.5523215	10
2	5.7954123	4.1779461	10
Total	6.0660993	4.2616726	20

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	1.46542938	1	1.46542938	0.08	0.7849
Within groups	343.609781	18	19.0894323		
Total	345.07521	19	18.1618532		

Bartlett's test for equal variances: $\chi^2(1) = 0.0627$ Prob> $\chi^2 = 0.802$

. *****

. oneway length treatment if time==0 & type== "pullet", tabulate

treatment	Summary of length		
	Mean	Std. Dev.	Freq.
1	1321.6	138.83191	5
2	1118.4	53.891558	5
Total	1220	146.03653	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	103225.6	1	103225.6	9.31	0.0158
Within groups	88714.4	8	11089.3		
Total	191940	9	21326.6667		

Bartlett's test for equal variances: $\chi^2(1) = 2.7982$ Prob> $\chi^2 = 0.094$

. oneway length treatment if time==0 & type== "ga_de", tabulate

treatment	Summary of length		
	Mean	Std. Dev.	Freq.
1	1347	53.744767	5
2	1259	319.43779	5
Total	1303	220.87603	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	19360	1	19360	0.37	0.5604
Within groups	419716	8	52464.5		
Total	439076	9	48786.2222		

Bartlett's test for equal variances: $\chi^2(1) = 7.9437$ Prob> $\chi^2 = 0.005$

. oneway length treatment if time==0 , tabulate

treatment	Summary of length		
	Mean	Std. Dev.	Freq.
1	1334.3	100.14661	10
2	1188.7	228.32727	10

Total | 1261.5 187.1477 20

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	105996.8	1	105996.8	3.41	0.0813
Within groups	559464.2	18	31081.3444		
Total	665461	19	35024.2632		

Bartlett's test for equal variances: chi2(1) = 5.2343 Prob>chi2 = 0.022

. oneway length treatment if time==1 & type== "pullet", tabulate

Summary of length			
treatment	Mean	Std. Dev.	Freq.
1	1175.4	278.4552	5
2	1323	93.525398	5
Total	1249.2	210.7135	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	54464.4	1	54464.4	1.26	0.2938
Within groups	345137.2	8	43142.15		
Total	399601.6	9	44400.1778		

Bartlett's test for equal variances: chi2(1) = 3.5894 Prob>chi2 = 0.058

. oneway length treatment if time==1 & type== "ga_de", tabulate

Summary of length			
treatment	Mean	Std. Dev.	Freq.
1	1257.8	110.89274	5
2	1240.2	160.46713	5
Total	1249	130.36786	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	774.4	1	774.4	0.04	0.8451
Within groups	152187.6	8	19023.45		
Total	152962	9	16995.7778		

Bartlett's test for equal variances: chi2(1) = 0.4748 Prob>chi2 = 0.491

. oneway length treatment if time==1 , tabulate

Summary of length			
treatment	Mean	Std. Dev.	Freq.
1	1216.6	204.48102	10
2	1281.6	131.28696	10
Total	1249.1	170.53535	20

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	21125	1	21125	0.72	0.4087
Within groups	531438.8	18	29524.3778		
Total	552563.8	19	29082.3053		

Bartlett's test for equal variances: chi2(1) = 1.6219 Prob>chi2 = 0.203

. oneway deepth treatment if time==0 & type== "pullet", tabulate

Summary of deepth			
treatment	Mean	Std. Dev.	Freq.

1	264.6	30.492622	5
2	201.2	27.471804	5
Total	232.9	43.18809	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	10048.9	1	10048.9	11.93	0.0086
Within groups	6738	8	842.25		
Total	16786.9	9	1865.21111		

Bartlett's test for equal variances: $\chi^2(1) = 0.0386$ Prob> $\chi^2 = 0.844$

. oneway depth treatment if time==0 & type== "ga_de", tabulate

Summary of depth			
treatment	Mean	Std. Dev.	Freq.
1	299	38.255719	5
2	201.8	61.140821	5
Total	250.4	70.25857	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	23619.6	1	23619.6	9.08	0.0167
Within groups	20806.8	8	2600.85		
Total	44426.4	9	4936.26667		

Bartlett's test for equal variances: $\chi^2(1) = 0.7546$ Prob> $\chi^2 = 0.385$

. oneway depth treatment if time==0 , tabulate

Summary of depth			
treatment	Mean	Std. Dev.	Freq.
1	281.8	37.314876	10
2	201.5	44.687184	10
Total	241.65	57.466031	20

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	32240.45	1	32240.45	19.02	0.0004
Within groups	30504.1	18	1694.67222		
Total	62744.55	19	3302.34474		

Bartlett's test for equal variances: $\chi^2(1) = 0.2757$ Prob> $\chi^2 = 0.600$

. oneway depth treatment if time==1 & type== "pullet", tabulate

Summary of depth			
treatment	Mean	Std. Dev.	Freq.
1	239	62.437969	5
2	265.8	146.56637	5
Total	252.4	107.1429	10

Analysis of Variance					
Source	SS	df	MS	F	Prob > F
Between groups	1795.6	1	1795.6	0.14	0.7166
Within groups	101520.8	8	12690.1		
Total	103316.4	9	11479.6		

Bartlett's test for equal variances: $\chi^2(1) = 2.3248$ Prob> $\chi^2 = 0.127$

```
. oneway depth treatment if time==1 & type== "ga_de", tabulate
```

treatment	Summary of depth		
	Mean	Std. Dev.	Freq.
1	272	80.177927	5
2	207.4	36.882245	5
Total	239.7	67.977202	10

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	10432.9	1	10432.9	2.68	0.1403
Within groups	31155.2	8	3894.4		
Total	41588.1	9	4620.9		

Bartlett's test for equal variances: $\chi^2(1) = 1.9578$ Prob> $\chi^2 = 0.162$

```
. oneway depth treatment if time==1 , tabulate
```

treatment	Summary of depth		
	Mean	Std. Dev.	Freq.
1	255.5	69.94482	10
2	236.6	105.35358	10
Total	246.05	87.572812	20

Source	Analysis of Variance			F	Prob > F
	SS	df	MS		
Between groups	1786.05	1	1786.05	0.22	0.6422
Within groups	143924.9	18	7995.82778		
Total	145710.95	19	7668.99737		

Bartlett's test for equal variances: $\chi^2(1) = 1.3923$ Prob> $\chi^2 = 0.238$

```
.  
end of do-file
```

```
. log close  
  name: <unnamed>  
  log: D:\workplace\project\NL group\PROBYN - BRENNTAG\moi\Untitled.log  
  log type: text  
  closed on: 2 Jul 2019, 16:36:58
```